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# The Effects of Temperature and Solids Retention Time on Activated Sludge Treatment Performance

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To the Graduate Council:

I am submitting herewith a thesis written by Jack McClendon Carley entitled "The Effects of Temperature and Solids Retention Time on Activated Sludge Treatment Performance." I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Environmental Engineering.

Dr. Kevin G. Robinson, Major Professor

We have read this thesis and recommend its acceptance:

Dr. Gregory Reed, Dr. Chris Cox

Accepted for the Council:

Carolyn R. Hodges

Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

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Dr. Chris Cox

Accepted for the Council:

Dr. Anne Mayhew  
Vice Provost and Dean of Graduate Studies

(Original signatures are on file with official student records.)

**The Effects of Temperature and Solids Retention Time on  
Activated Sludge Treatment Performance**

A Thesis

Presented for the

Master of Science Degree

The University of Tennessee, Knoxville

Jack McClendon Carley

December 2003

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## **Abstract**

A bench-scale treatment system with external clarifier was used to determine the effects of solids retention time (SRT) and temperature on treatment performance while simulating the full scale activated sludge process. Four bench-scale reactors, operating at SRTs of 20, 10, 5 and 2 days, were fed settled municipal wastewater collected after primary clarification from the Kuwahee wastewater treatment plant in Knoxville, TN. Three operational temperatures were investigated during this study, 22°C, 12°C and 7°C. The order and duration of operation at each temperature was first 22°C for 61 days, next 7°C for 47 days and lastly 12°C for 105 days.

Experimental results indicate that carbon treatment performance improved, on average with increasing SRT; however, carbon treatment performance was not effected by operational temperatures studied to the same extent. On average, carbon oxidation improved with increasing SRT at each operational temperature; however, carbon oxidation was more efficient at 12°C than 22°C. COD<sub>T</sub> treatment performance at 7°C was poor irrespective of SRT. The difference in performance between 22°C and 12°C was attributed to acclimation of biomass from startup at 22°C to operation at 12°C beginning at 108 days. Sludge volume index (SVI) and effluent suspended solids (ESS) values also indicated that SRT had a greater impact on biosolids settling than did temperature.

During this study, nitrification was observed to be viable at 22°C and 12°C despite reported sensitivity of lower SRTs (2 and 5 days) to process changes and low operational

temperature. According to these observations, higher SRTs would be recommended for full-scale treatment especially during wintertime or sub-optimum temperatures. Nitrification at 7°C was observed to be almost nonexistent for all SRTs studied during this research. Based on the data presented in this study, an SRT of 5 days and a process temperature of 12°C are necessary to achieve effective carbon treatment, biosolids settling, and nitrification.

A comparison of the full and bench scale (5-day SRT) systems, based on the operational and environmental parameters considered in this study revealed inadequacies of the bench scale system in simulating the full scale facility. Carbon treatment performance as well as MLSS levels, differed in the two systems. The full scale facility consistently discharged significantly lower carbon levels than did the bench scale system. Additionally MLSS levels were significantly higher in the full scale facility. Nitrification performance was better in the bench scale system at temperatures above 12°C but were significantly poorer at 7°C. Overall, the large variability in the performance data obtained for the two systems indicated that the bench scale system did not effectively simulate the full scale facility.

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# Nomenclature and Abbreviations

## Nomenclature

$b$	Specific Biomass Decay Rate
$pH$	Hydrogen Ion Concentration
$q$	Specific Substrate Utilization Rate
$q_{\max}$	Maximum Specific Substrate Utilization Rate
$Q$	Flow Rate
$Q_r$	Recycle Flow Rate
$Q_w$	Wastage Flow Rate
$S$	Rate-limiting Substrate Concentration
$S_o$	Influent Substrate Concentration
$S_e$	Effluent Substrate Concentration
$T$	Temperature (Absolute or Centigrade)
$V$	Volume
$X$	Cell Mass Concentration
$X_e$	Cell Mass Concentration in Effluent
$Y$	Yield Coefficient
$\theta$ or $\theta_N$	Thermal Coefficient
$\theta_c$	Solids Retention Time
$\theta_c^d$	Design Biological Solids Retention Time
$\theta_c^{\min}$	Minimum Biological Solids Retention Time to Ensure Nitrification
$\mu$	Specific Growth Rate
$\mu_{\max}$	Maximum Specific Growth Rate
$\mu_{N,\max}$	Maximum Specific Nitrifier Growth Rate

## Abbreviations

ASCE	American Society of Civil Engineers
BOD	Biological Oxygen Demand
BOD <sub>5</sub>	Five Day Biological Oxygen Demand
COD	Chemical Oxygen Demand
COD <sub>T</sub>	Total Chemical Oxygen Demand
DO	Dissolved Oxygen
ESS	Effluent Suspended Solids
F: M	Food to Microorganism Ratio
HDPE	High Density Polyethylene
HRT	Hydraulic Retention Time
HVAC	Heating Ventilation Air Conditioning
IC	Ion Chromatograph
i.d.	Inside Diameter
LDL	Lower Detection Limit

MGD	Million Gallons Per Day
MLSS	Mixed Liquor Suspended Solids
MLVSS	Mixed Liquor Volatile Suspended Solids
MST	Maximum Storage Time
NOB	Nitrite Oxidizing Bacteria
NOD	Nitrogenous Oxygen Demand
NPT	National Pipe Thread
O.D.	Outside Diameter
RRNA	Ribosomal Ribonucleic Acid
SBR	Sequencing Batch Reactor
SRT	Solids Retention Time
SVI	Sludge Volume Index
TKN	Total Kjeldahl Nitrogen
USEPA	United States Environmental Protection Agency
VSS	Volatile Suspended Solids
WEF	Water Environment Federation
WWTP	Wastewater Treatment Plant

# Chapter 1.0

## Introduction

Many different treatment strategies are employed for the removal of biochemical oxygen demand (BOD) and conversion of organic nitrogen (N) to nitrate from municipal wastewater before it is discharged to receiving waters. Bacteria, cultivated in these treatment processes, perform biological functions that utilize BOD and organic nitrogen consequently removing the target pollutants (BOD) or converting them to more innocuous forms (i.e. nitrogen). Environmental engineers use these microbial mediated processes as tools to remediate target pollutants from aquatic environments.

Organic carbon within a wastestream creates an oxygen demand when discharged into receiving waters. Biochemical oxygen demand is the quantity of oxygen used by microorganisms in the aerobic stabilization of wastewaters and polluted waters. In wastewater engineering, BOD is frequently used to define influent and effluent characteristics and to assess treatment process efficiency (Aziz and Tebbutt, 1979).

Additionally, nitrogenous compounds are important in wastewater management due to the detrimental impact of these compounds on aquatic environments (Yantarasri *et al.* 1992). For example, ammonia-nitrogen ( $\text{NH}_4^+\text{-N}$ ) discharged into receiving streams can result in oxygen depletion, eutrophication, and toxicity to aquatic life forms (USEPA 1993). Microbial utilization of ammonia is a process whereby this compound is removed from wastewater streams. Nitrification is the biological process whereby free and saline ammonia is oxidized to nitrite ( $\text{NO}_2^-$ ) and then nitrate ( $\text{NO}_3^-$ ) (Ekama *et al.* 1984).

Nitrifying bacteria are used to ameliorate potential anthropogenic damage to aquatic environments through reduction of ammonia in wastewater during sewage treatment.

In a typical municipal wastewater environment, ammonia can far exceed the nutritional demands of microorganisms in biotreatment reactors. Activated sludge is one of the most common types of treatment techniques used in the removal of ammonia-nitrogen and BOD from a municipal waste stream (ASCE Manual No. 76). A typical activated sludge system is an aerobic, suspended-growth, continuous flow reactor with an external clarifier; although, many process variations exist. External clarification allows for recycle of settled biomass from reactor effluent, which can improve treatment performance by increasing the concentration of biosolids in the reactor. The efficiency of biologically mediated processes, such as the ones supported in activated sludge, are dependent on certain operational and environmental factors, such as solids retention time (SRT), temperature, pH, dissolved oxygen (DO) and alkalinity. Hence the efficacy of this wastewater treatment process is dependent on these parameters.

Solids retention time (SRT) plays a large part in carbon and nitrogen removal and subsequent process control. Solids retention time is defined as the average amount of time that bacteria are present within the reactor. This amount of time is controlled by removing a calculated amount of biomass (typically new growth) from the reactor contents. Subsequently, treatment performance can be modified by altering biomass removal rates and therefore SRT.

Temperature also plays a very important role in biological treatment. The temperature to which the nitrifying bacteria are exposed affects the metabolic processes involved in nitrification and the rate of growth of those bacteria. Cold temperature and



its negative effect on reaction rates become an important factor in ensuring efficient ammonia removal performance in wastewater treatment. Since temperature can not be controlled practically, quantification of the effect of temperature allows engineers to make process modifications to provide consistent and efficient level of treatment under less than optimal conditions.

This study was part of a larger research project whose overall goal was the comparison of traditional physio-chemical methods and molecular techniques to assess treatment performance. Such a comparison would allow for correlation of bacterial numbers with treatment performance data. This bench scale study attempted to simulate a local, full scale municipal WWTP through the assessment of activated sludge treatment performance using traditional physio-chemical methods of analysis while investigating temperature and SRT as the main operational and environmental parameters. The specific objectives of this study include:

1. to develop and optimize a bench-scale reactor/external clarifier (with biosolids recycle) that simulates the nitrifying activated sludge process used in the local full-scale municipal WWTP, and
2. to evaluate bench-scale performance relative to full-scale operation and assess the effectiveness of the bench-scale system in simulating the full-scale process, and
3. to determine the impact of a major operational parameter (SRT), and a major environmental parameter (temperature), on carbon and nitrogen removal.

## **Chapter 2.0**

### **Literature Review**

#### **2.1 Conventional Activated Sludge Treatment Process**

Bacteria have long been employed in wastewater treatment to remove organic carbon and ammonia. There are two basic options in biological wastewater treatment design, attached or suspended growth systems, which promote the growth of bacteria involved in removal of anthropogenic compounds present in municipal wastewater. The most widely used treatment process for biological removal of nitrogenous and carbonaceous compounds is the complete mix activated sludge suspended growth process (Figure 1). In this process, the introduction of a continuous flow of wastewater is rapidly distributed, due to vigorous mixing, throughout the aeration basin. Biological treatment occurs while wastewater is in contact with microorganisms contained within activated sludge. Separation of solids containing required bacteria for treatment from the wastewater stream is achieved through clarification. Clarification is typically achieved through gravity separation of flocculated solids from the treated wastewater.

Typical activated sludge design includes the use of a secondary clarifier. The secondary clarifier offers several advantages in treatment operation. First, the clarifier provides a means to concentrate and collect the mixed-liquor suspended solids (MLSS) and recycle them back to the aeration chamber, thereby increasing the contact time between microorganisms and the wastewater. This introduces the concept of solids retention time (SRT) also denoted as sludge age ( $\theta_c$ ).

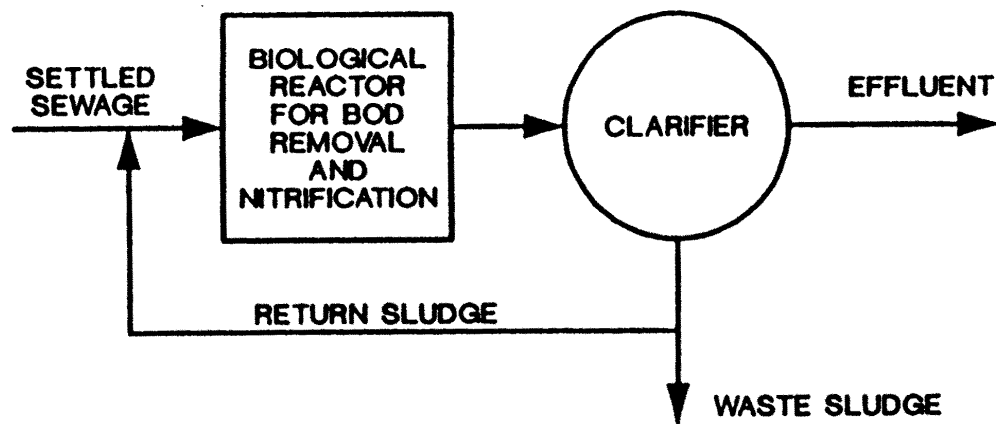


Figure 1. Process diagram for complete mix activated sludge treatment system.

Simply put, SRT is the average amount of time the bacteria exist within the aeration chamber. Lawrence and McCarty (1970) identify SRT as a central variable in the development of a unified basis for biological treatment design and operation. SRT is felt to be a particularly useful parameter because of its basic relationship to bacterial growth rate and the relative ease with which it can be used in design calculations and in the control of biological treatment systems (Lawrence & McCarty, 1970). For an activated sludge system without a secondary clarifier, the hydraulic retention time (HRT) would be equal to the SRT. With solids recycle, SRTs can be achieved which exceed the HRT of the system. Therefore, solids recycle allows for the decoupling of SRT from HRT, providing control of the residence time for the solids within the treatment system. In a complete mix activated sludge system that allows for solids recycle, time of contact between bacteria and wastewater is increased. This provides an increased chance that

the target compound(s) will be remediated, and therefore increasing the efficacy of the treatment system.

Secondly, the concentrated biomass in the 'underflow' (i.e. the recycle flow of settled solids) can be a suitable location to remove excess biomass in order to control SRT. Wastage of solids is necessary in a recycle system to avoid a build-up of biomass in the reactor and clarifier. Wastage of solids is also the means to control SRT. SRT further defined is the total amount of biomass in the treatment system divided by the total amount of biomass lost or wasted daily (Lawrence & McCarty, 1970). While longer SRTs provide more efficient treatment in general, maintenance of high solids concentrations can adversely affect the separation of solids from treated effluent in the clarifier resulting in an increase of effluent suspended solids. Calculated control of waste solids, and therefore SRT, for complete mix activated sludge is one of the more useful tools wastewater engineers employ to ensure efficient wastewater treatment.

The SRT of the reactor system is directly related to the growth rate of the microorganisms involved in the treatment process (Lawrence & McCarty, 1970). With constant conditions, such as available substrate, pH, dissolved oxygen (DO) and temperature, the operation of a complete mix activated sludge system will arrive at steady state. Steady state is an operational condition that exists when the rate of bacterial growth and the rate of solids removal are equal, resulting in a constant concentration of biomass in the aeration chamber over time. This operational characteristic for complete mix activated sludge is necessary to make simplifying assumptions when determining kinetic coefficients.

It is also important to mention that secondary clarification can influence the microbial community structure present in the complete mix activated sludge system. Secondary clarification takes advantage of the settling characteristics of the floc (i.e. a conglomerate of bacteria). Flocculating bacteria have a selective advantage over non-flocculating bacteria for several reasons. Settable flocs will be returned to the reactor through gravity separation, whereas freely growing bacteria tend to remain in suspension, and will ultimately be lost in the effluent; thus, selecting for flocculating bacteria. Flocculation can also provide protection for bacteria from natural predators, such as protozoa, since these organisms mainly feed on freely growing cells (Eckenfelder et al, 1998).

## **2.2 Municipal Wastewater Characteristics**

Municipal wastewater has general characteristics inherent due to the anthropogenic origin of this waste stream. The components that make up the wastewater from a community depend on the type of collection system used and may include domestic wastewater, industrial wastewater, infiltration and storm water (Metcalf and Eddy, 1981). Metcalf and Eddy (1991) present typical U.S. wastewater characteristics based on the general strength of the wastewater (Table 1). These typical wastewater characteristics agree with other sources (Tchobanoglous & Burton, 1991 and WEF Manual of Practice No. 8, 1991) containing similar categorical information. As can be seen from Table 1, the values for BOD<sub>5</sub> and COD for typical U.S. municipal wastewater range between 110 to 400 mg/L and 250 to 1000 mg/L, respectively. The nondegradable fraction of the oxygen demand is an important factor when defining treatment. Viessman and Hammer (1991) report a refractory portion of organic compounds range from 20 to

40%. Ekama *et al.* (1984) report that the unbiodegradable particulate COD fraction is less than 10% for settled wastewater.

The nitrogen speciation presented in Table 1 shows total nitrogen ranging between 20 and 85 mg/L where ammonia-nitrogen comprises approximately 60% of the total leaving 40% as organically bound nitrogen. The presence of nitrite ( $\text{NO}_2^-$ ) and nitrate ( $\text{NO}_3^-$ ) in untreated domestic wastewater is insignificant with respect to the total nitrogen concentration. McCarty (1970) reported  $\text{NO}_2^-$  and  $\text{NO}_3^-$  concentrations in raw wastewater ranging from 0 to 0.1 mg/L and 0 to 0.5 mg/L, respectively.

Table 1. General U.S. municipal wastewater characteristics.

Item	Concentration	
	Weak	Medium
Biochemical Oxygen Demand ( $\text{BOD}_5$ ) mg/L	110	220
Chemical Oxygen Demand (COD), mg/L	250	500
Suspended Solids		
Total	100	220
Volatile	80	165
Nitrogen (as N), mg/L		
Organic	8	15
Ammonia	12	25
Total	20	40
Phosphorus (as P) mg/L		
Organic	1	3
Inorganic	3	5
Total	4	8

\* adapted from Metcalf and Eddy (1991)

## 2.3 Nitrification Process Overview

### 2.3.1 Microbiology of Nitrification

Activated sludge should be understood as an artificial living ecosystem, which is under continuous influence of abiotic and biotic factors (Eckenfelder *et al.*, 1998). The general health of the biomass contained within the activated sludge process is dependent on the interactions between populations of bacteria present and other environmental factors, including but not exclusive to temperature, substrate concentration, pH and dissolved oxygen concentration. According to Eckenfelder *et al.* (1998), activated sludge microorganisms can be segregated into two major groups, decomposers and consumers. Decomposers are responsible for biochemical degradation of polluting substances in wastewater, and include bacteria, fungi and colorless cyanophyta. Consumers utilize bacterial and other microbial cells as a substrate; this segment of the activated sludge population includes predator type organisms such as phagotrophic protozoa and microscopic metazoa. Nitrifying bacteria are part of the decomposer class of microorganisms present in activated sludge.

There are two distinct, physiologically defined groups of bacteria that catalyze the two half-reactions (ammonia and nitrite oxidation) involved in nitrification (Bock *et al.*, 1992). Nitrifying bacteria are classified as chemolithoautotrophs, a group of bacteria that derive cellular energy from oxidation of inorganic nitrogen. Chemolithoautotrophs assimilate inorganic carbon, present as alkalinity in aquatic systems, in the form of new cells and cellular compounds. Sixteen species of lithoautotrophic ammonia-oxidizing bacteria have been isolated and described (Jones *et al.*, 1998).

Ammonia oxidizers comprise two groups within the class of *Proteobacteria*. *Nitrosococcus oceanus* and *N. halophilus* belong to the gamma (γ) subclass of the class *Proteobacteria* (Woese *et al.*, 1984), while the members of the genera *Nitrosomonas*, *Nitrospira*, *Nitrosovibrio*, and *Nitrosolobus*, as well as *Nitrosococcus mobilis* constitute a closely related assemblage within the beta subclass of *Proteobacteria* (Pommerening-Roser *et al.*, 1996). Based on ultra-structural properties, cultivated nitrite-oxidizing bacteria have been assigned to the four recognized genera *Nitrobacter*, *Nitrospina*, *Nitrococcus*, and *Nitrospira* (Juretschko *et al.*, 1998). The specific genera of ammonia and nitrite oxidizing bacteria present is dependent on the environmental locus (Prosser, 1989).

Investigation of the diversity and ecology of nitrifying bacteria in natural and engineered systems by traditional cultivation techniques has been hampered by slow bacterial growth and by the biases inherent in culture-based studies (Wagner *et al.*, 1993). Mobarry *et al.* (1996) studied nitrifying microbial community populations in activated sludge and biofilm systems using 16s rRNA targeted DNA probes for phylogenetically defined groups of autotrophic ammonia- (AOB) and nitrite-oxidizing (NOB) bacteria. These experiments revealed that *Nitrosomonas* and *Nitrobacter* species occurred in clusters (i.e. flocs) and frequently were in contact within heterotrophic sludge flocs (Mobarry *et al.*, 1996). Also very little hybridization was observed with the *Nitrobacter* and *Nitrosovibrio-Nitrospira-Nitrosolobus* probes, indicating that their contribution to nitrification in this system was minor or that there were uncharacterized AOB present. Quantitative hybridization with oligonucleotide probes have shown that, in ammonia-rich systems like activated sludge or biofilm reactors, the dominant AOB were members of

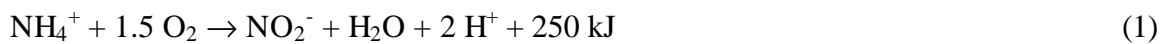


the beta ( $\beta$ ) subgroup of the *Proteobacteria* and, more specifically, *Nitrosomonas* species (Roberts and Lewis, 2001).

### 2.3.2 Energy and Synthesis Pathways

Nitrification is a chemolithotrophic process where nitrifying bacteria utilize energy generated from the oxidation of inorganic nitrogen (Eckenfelder and Grau, 1998). Figure 2 shows the oxidation states of various nitrogenous compounds. Lower values of oxidation states correlates with more highly reduced compounds. Reduced compounds have electrons that are released during oxidation in the form of hydrogen ions.. These electrons are captured during bacterial catabolism as  $\text{NADH}+\text{H}^+$  and  $\text{FADH}_2$  for use in oxidative phosphorylation (Shuler, 1992). The greater the reduction-oxidation potential between substrate and end product, the more energy is available through the oxidation of that compound.

As described earlier, nitrification is performed in two distinct processes by different kinds of chemolithotrophic microorganisms, ammonia oxidizing bacteria (AOB) and nitrite oxidizing bacteria (NOB). During nitrification the oxidation state of nitrogen incrementally changes from  $-3$  to  $+5$ , as indicated by Figure 2. The two-step process of nitrification can be described by the following stoichiometric relationships:



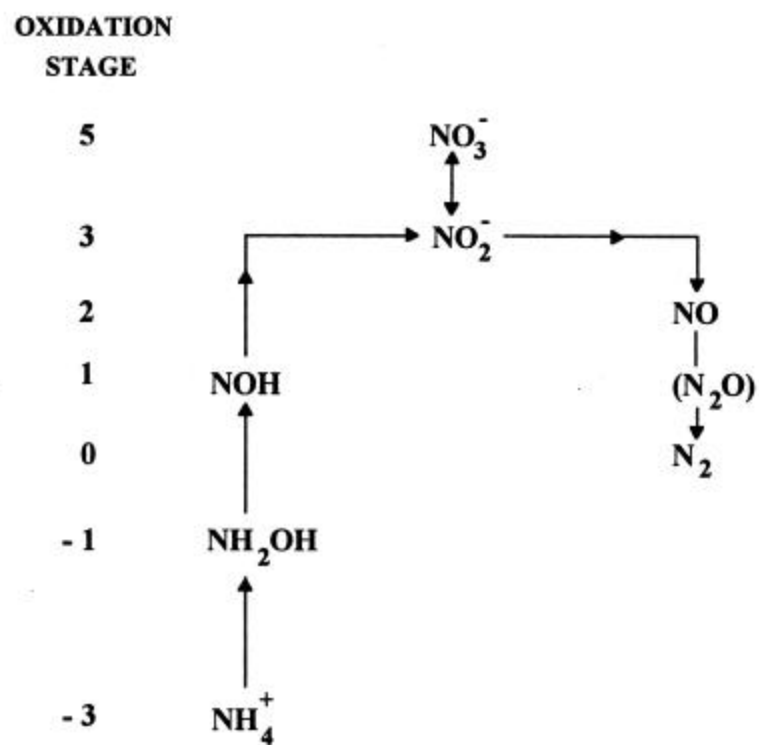


Figure 2. Oxidation stages of nitrogen compounds in wastewater treatment  
\* adapted from Barnes and Bliss (1983)

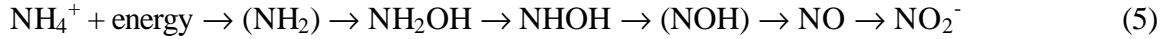
Equation 1 and Equation 2 represent the separate processes facilitated by (1) AOB and (2) NOB. The free energy change ( $\Delta G$ ) for the two reactions is  $-250$  kJ and  $-75$  kJ, respectively. From these  $\Delta G$  values, the first step in nitrification (ammonia oxidation) yields approximately three times the amount of free energy compared to the second step (nitrite oxidation). This is supported by the change in oxidation state between the product and reactant for the two steps. The first step provides a change in oxidation state from  $-3$  to  $+3$ , whereas the second step provides a change in oxidation state from  $+3$  to  $+5$ . The oxidation-reduction potential during the conversion of ammonia to nitrite is three times greater as compared to the oxidation of nitrite to nitrate. The difference in oxidation-reduction potential and  $\Delta G$  are relatively similar in comparison of both steps. Both the reduction-oxidation potential and free energy change illustrate that ammonia oxidation utilizes the majority of available energy from the overall process of nitrification.

The stoichiometric description of the first step of nitrification, oxidation of ammonia to nitrite is somewhat of an over simplification. This bioprocess is a summation of two coupled biological reactions (Equations 3 and Equation 4).



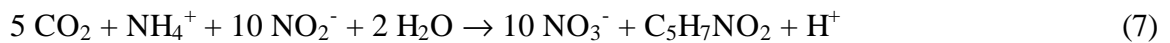
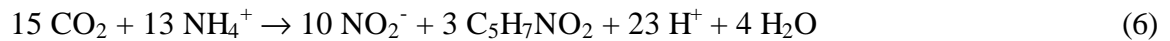
These two equations illustrate the conversion of ammonia to an intermediate compound, hydroxylamine ( $\text{NH}_2\text{OH}$ ). Energetically, it is necessary to couple these reactions due to the fact that Equation 3 is endergonic and energy is produced only in the

following step (i.e. the oxidation of hydroxylamine to nitrite) (Eckenfelder and Grau, 1998). Due to the complexity of ammonia oxidation to nitrite, the pathway by which *Nitrosomonas* metabolizes ammonia to nitrite is still speculative (Grady *et al.*, 1980). An alternate hypothesis proposed by Grady *et al.* (1980) is presented as Equation 5.

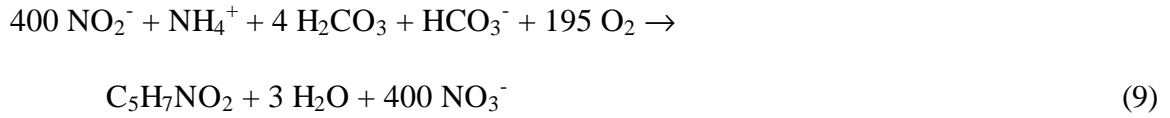
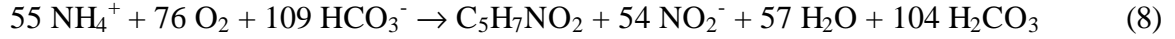


The compounds in parenthesis are thought to be free radical-like intermediates, which are bound to an enzyme (Grady *et al.*, 1980).

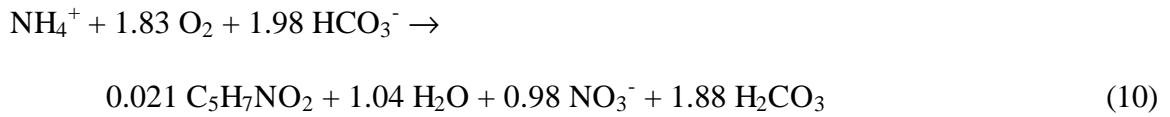
If it is assumed that the empirical formula for the elemental composition of a bacterial cell is  $\text{C}_5\text{H}_7\text{NO}_2$ , stoichiometric relationships can be derived for theoretical cell growth for AOB and NOB (Equation 6 and Equation 7) (Gujer *et al.*, 1974; USEPA, 1993).



Through the combination of energy and synthesis equations presented previously, overall synthesis-oxidation equations can be concluded using representative measurements of yields and oxygen consumption for AOB and NOB. The overall synthesis-oxidation relationships are presented by Equation 8 and Equation 9 for *Nitrosomonas* and *Nitrobacter*, respectively (USEPA, 1975).



The overall synthesis-oxidation reaction for the entire nitrification process is presented by Equation 10 (USEPA, 1993).



In these equations, yields for *Nitrosomonas* and *Nitrobacter* are 0.15 mg cells / mg  $\text{NH}_4^+$ -N and 0.02 mg cells / mg  $\text{NO}_2^-$ -N, respectively. On this basis, the removal of 20 mg/L of ammonia nitrogen would yield only 1.8 mg/L of nitrifying organisms (USEPA, 1993). Such low yields can prove to be problematic when adverse conditions limit bacterial metabolism. These low biomass yields provide, in part, a basis for sensitivity of the nitrification process.

## 2.4 Environmental Effects on Nitrification and other Biochemical Unit Processes

### 2.4.1 Temperature

It is a well-known fact that nitrification is a temperature-sensitive process that can be affected by seasonal temperature variations (Harremoes *et al.*, 1998; Sinkjaer *et al.*, 1996; USEPA, 1993; Tendaj *et al.*, 1992; Ford, 1980). Knowledge of the favorable range

for some environmental parameters, i.e., dissolved oxygen (DO), pH, etc., may be utilized directly in optimizing a treatment process. However, seasonal variation of temperature is not an environmental factor that can be directly controlled in most activated sludge systems (Sharma *et al.*, 1976).

Bacteria generally flourish in an environment, which meets certain prerequisites for growth. Temperature is one of those prerequisites. Autotrophic and heterotrophic bacteria involved in biological wastewater treatment can be generally classified, based on growth as a function of temperature, as mesophilic. This stems from the origin of the bacteria, which are from the digestive tract of humans (heterotrophic) and other soil-borne bacteria (autotrophs). Mesophilic bacteria grow best in temperate environments between 20 and 50°C, as compared to psychrophiles and thermophiles, which prefer colder (<20°C) or hotter (>50°C) conditions, respectively (Shuler, 1992). However the nitrification process has been observed over a range of temperatures from approximately 4°C to 45°C (USEPA, 1993). The overall optimum temperature reported for growth of nitrifying bacteria appears to be in the range of 28 - 36°C with some variation in reported range of values. The Manual for Nitrogen Control published by the USEPA (1993) reported a temperature growth optimum for *Nitrosomonas* as being about 35°C and an optimum growth range for *Nitrobacter* as 35 – 42°C. These values were originally presented by Buswell *et al.*, (1954); Deppe and Engle, (1960) and Laudelout and Van Tichelen, (1960). Grunditz and Dalhammar (2001) isolated and characterized, with respect to temperature, *Nitrosomonas* and *Nitrobacter* from a nitrifying activated sludge from a large wastewater treatment plant in Stockholm. The test organisms were characterized with respect to temperature. The highest activity was found at a

temperature of 35°C for *Nitrosomonas* and at 38°C for *Nitrobacter*. These values agree with the optimum temperature ranges for nitrification presented by the U.S. EPA, (1993).

Full-scale WWTPs are exposed to temperature variation from seasonal changes, wind exposure and influent wastewater (Jansen *et. al.*, 1992). In northern climates, significantly greater SRTs are required for complete nitrification in winter than are necessary during summer (Sharma *et. al.*, 1976). This illustrates the point that cold process temperatures are unavoidable or even commonplace for some treatment plants, thus constituting an operational difficulty. Understanding temperature effects on biological wastewater treatment, environmental engineers can avoid potential process failure through appropriate design and operational changes. Correcting kinetic constants for low or sub-optimum process temperatures redefines the minimum operating SRT ( $\theta_c^{\min}$ ) and subsequently the design SRT ( $\theta_c^d$ ). There is a pronounced decrease in nitrification at temperatures below 18°C with little nitrifier growth occurring below 5°C (Ford, 1980). Conversely, at higher temperatures Barritt (1933) found that the thermal death point of a pure culture of *Nitrosomonas* was between 54 and 58°C.

#### **2.4.2 Effect of Temperature on Nitrification**

It has been shown that temperature effects the kinetics of nitrifier growth and substrate utilization rates. There are five main kinetic parameters which describe a microbial population. These kinetic parameters are the maximum specific growth rate ( $\mu_{\max}$ ), maximum specific substrate utilization rate ( $q_{\max}$ ), saturation constant ( $K_s$ ), the yield coefficient ( $Y$ ) and the microbial decay rate ( $b$ ). The following discussion reviews

literature findings on the effect of temperature on these kinetic coefficients with respect to nitrification.

*Maximum specific growth rate,  $\mu_{N,max}$*

The maximum specific growth rate is a commonly reported parameter in quantification of microbial growth. In some of the earliest studies observing temperature effects on nitrification, Downing *et al.* (1964) reported that the growth rate constant roughly doubles for each 10°C increase in temperature in the range of 7°C to 25°C. Knowles *et al.* (1965) observed an increase in maximum specific growth rate for *Nitrosomonas* was 9.5% per degree Centigrade, and 5.9% increase per degree Centigrade for *Nitrobacter*. Srna & Baggaley (1975) measured the effect of rapid temperature changes on nitrification reactions via batch studies on a marine nitrifying filter system. The results revealed that a 4°C increase in temperature caused about a 50% increase in the rate of nitrification, and a 1°C drop caused a 30% decrease when compared with calculated values at 21.3°C. Jenkins *et al.* (1967) reported a 3% increase in growth rate for a 10°C increase in temperature. In a laboratory experiment, Painter and Loveless (1983) observed a 4.8% increase in the rate of nitrification per degree Centigrade. Sozen *et al.* (1996) reported that net growth for nitrifiers was highly variable within a range of 0.24-0.52 d<sup>-1</sup>, with an average value of 0.38 d<sup>-1</sup>. At 10°C, the net growth rate was observed to drop to 0.14 d<sup>-1</sup>, ranging from 0.10-0.17 d<sup>-1</sup>, which showed less variation than at 20°C.

Considering a practical temperature range (< 30°C), temperature optima for growth are rarely encountered in municipal wastewater treatment. However operation at



cold temperatures result in slow growth rates of nitrifying bacteria hindering treatment performance especially with respect to ammonia oxidation.

*Maximum specific substrate utilization rate,  $q_{N,max}$*

There is a direct relationship between the maximum specific substrate utilization rate and the maximum rate of growth. This equation represents a linear relationship between these two maximum rates via a proportionality constant (the growth yield coefficient). Therefore theoretically  $q_{max}$  and  $\mu_{max}$  should have the same response to temperature. Other references also documented suppressed rates of ammonia oxidation with decline in temperature (Stark, 1996; Jansen, 1992; Wong-Chong *et al.*, 1978).

For heterotrophic activity, the maximum substrate utilization rate was found to increase with increases in temperature in both aerobic and anaerobic biological systems (Novak 1974). Tchobanoglous and Schroeder (1985) suggest a theta ( $\theta$ ) value of 1.047 for BOD temperature corrections.

*Saturation Constant,  $K_S$*

Early work by Downing *et. al.* (1964) reported an equation from estimation of a trendline from a log-normal plot of  $K_S$  versus temperature. This research is included in USEPA Process Design Manual for Nitrogen Control (USEPA 1993).

Temperature correction for the saturation constant is not normally reported due to its assumed consistency over a practical range of temperatures; however, the modified Arrhenius temperature correction coefficient,  $\theta$ , derived from Equation 16 was found to be 1.12. In addition, in aerobic systems, heterotrophic  $K_S$  increased as temperature

increased, whereas, in anaerobic systems, heterotrophic  $K_S$  decreased with an increase in temperature (Mines 1999).

#### *Endogenous decay coefficient, $b$*

The endogenous decay coefficient for nitrification,  $b_N$ , was found to vary with temperature, approximately doubling with a  $10^\circ\text{C}$  increase in temperature (Mines and Sherrard, 1999). Also as indicated by some researchers (Argaman, 1995; Benefield and Randall, 1980), the endogenous decay coefficient for nitrification varied with temperature according to a modified Arrhenius equation over the temperature range 15 to  $25^\circ\text{C}$ .

The heterotrophic endogenous decay coefficient,  $b_H$ , was also seen to vary with temperature. Argaman (1995) presented a modified-Arrhenius equation for modeling the effect of temperature on  $b_H$  for single-sludge activated sludge model. Collins and Grady (1978) cited literature (Muck and Grady, 1974; Mennet and Nakayama, 1971; Topiwala and Sinclair, 1971) presented a similar increase in heterotrophic endogenous decay with increasing temperature. A graph of microbial decay versus temperature for literature cited by Collins and Grady (1978) illustrated a 1.5, 1.6 and 3.3 fold increase in the decay rate for a  $10^\circ\text{C}$  increase in temperature. The temperature response depicted by the graph was linear.

#### *Yield Coefficient, $Y$*

Since observed yields are related to the true growth yield by a function containing the decay rate and SRT the observed and subsequently the true growth yield will display a maximum value with respect to temperature.

Benefield and Randall (1980) presented data that showed the yield coefficient for nitrification is generally independent of temperature over the temperature range from 15 to 25°C. Collins and Grady (1978) compared four sets of data from literature pertaining to the temperature dependency of heterotrophic growth yield,  $Y_H$ . The data suggest that there is a maximum in growth yield near 20°C. Due to the discontinuous nature of growth yield over a practical temperature range (< 30°C) makes temperature correction using the Arrhenius equation impractical. Mines and Sherrard (1999) cited a literature review performed by Characklis and Gujer (1979) who reviewed heterotrophic yields from several literature sources. Randall *et al.* (1992) determined from bench-scale fully aerobic activated sludge systems that the yield coefficient, 0.41-mg VSS/ mg COD, was relatively constant over a temperature range of 10 – 20°C. This literature review indicated that the biomass yield is constant over a relatively wide range for many microorganisms and substrate.

## **2.5 Cold Temperature and Nitrification in Biological Wastewater**

### **Treatment**

Nitrifying bacteria are known to be sensitive to temperature (Barnes and Bliss, 1983). Since the rate of nitrification (which is strongly temperature dependent) typically governs design and operation of an activated sludge WWTP, and SRT which prevents nitrifier washout is required. This identifies the SRT as the main dimensioning parameter for treatment plants, and the cold and changeable winter temperatures at the plants constitute the most serious operational problem encountered (Jansen *et. al.*, 1992).

Jansen (1992) published observations on activated sludge nitrification at four Danish municipal wastewater treatment plants, documenting typical winter temperatures

of 5-10°C with values as low as 2°C and up to 9°C during the same winter. Additional temperature variations occurred from year to year. Jansen (1992) also notes that temperature variations are more likely to be explained by variations in the structure of the sewer system and catchment area, and the construction of the treatment plant.

Ilies and Mavinic (2001) studied the effect decreased ambient temperature on biological nitrification of high ammonia landfill leachate using a 4-stage Bardenpho process. They observed no effect on nitrification performance as the temperature was lowered from 20°C to 14°C incrementally. However, stable nitrification was lost when the temperature was lowered to 10°C resulting in nitrification efficiencies between 10 and 30% (Ilies and Mavinic, 2001). Nitrification inhibition was attributed primarily to the low operating temperature. Ilies and Mavinic (2001) also suggest that longer acclimatization periods for the nitrifying biomass coupled with smaller step decreases in temperature may be helpful in determining critical points for nitrification and suggest methods to prevent process failure. Rostron, Stuckey and Young (2001) also observed a small decrease (10%) in nitrification when process temperature was lowered from 25 to 16°C in a comparative study of immobilisation media to enhance nitrification of high strength ammonia wastewater.

Biological nitrogen removal at low temperatures was considered highly questionable, some sources reported that below 5°C the process of nitrification ceases, both in suspended growth (Anon, 1974) and in attached growth (Pano and Middlebrooks, 1983) systems. However Tendaj *et al.* (1992) reported that when temperature remains constant at a low level over a long period of time, nitrification gradually increases. These researchers, monitoring Swedish activated sludge systems, documented a sudden process

temperature drop from 14 to 12 °C. Due to the sudden drop in temperature the nitrification rate subsequently fell to 2.0 g N/kg VSS·h. During the two months following this drop in temperature, temperature remained constant at 12°C and the rate of nitrification rose to 2.7 g N/kgVSS·h. This 30% rate increase was due to the ability of the bacteria to adapt to a lower temperature (Tendaj *et.al.*, 1992). The rate after adjustment to 12°C was the same rate as that observed at 16°C.

Hoilijoki *et al.* (2000) studied the effect of low temperatures on nitrification for anaerobically pretreated landfill leachate using the activated sludge process. This study was designed to evaluate the use of carrier plastic as a means of ensuring process stability. The laboratory experiment without carrier plastic revealed at a loading rate of 0.027 g NH<sub>4</sub><sup>+</sup>-N MLVSS<sup>-1</sup> d<sup>-1</sup> and a HRT of 3 days, complete nitrification was observed at 10°C. At 7°C and a loading rate of 0.023 g NH<sub>4</sub><sup>+</sup>-N MLVSS<sup>-1</sup> d<sup>-1</sup> ammonia removal was only 93%. At 5°C and a loading rate of 0.011 g NH<sub>4</sub><sup>+</sup>-N MLVSS<sup>-1</sup> d<sup>-1</sup> only 61% ammonia removal was observed, while the activated sludge reactor with carrier plastic achieved complete ammonia removal.

Oleszkiewicz (1988) studied the effect of low temperatures on nitrification/denitrification using sequencing batch reactors (SBRs). This study revealed that the response to the decrease in temperature was discontinuous. Oleszkiewicz (1988) reported a more pronounced effect of temperature on nitrification from 2 – 7°C; due to this fact, a modified Arrhenius equation was used to describe both portions of the temperature curve. The temperature correction factors calculated on either side of the discontinuity (@ 7°C) were  $\theta_N = 1.02$  (15 to 7 °C) and  $\theta_N = 1.40$  (7 to 2 °C). However, Oleszkiewicz (1988) did document the feasibility of nitrification down to 2°C for SBRs

although steady 90% nitrification efficiency at 2°C could only be accomplished at SRT beyond 30 days accompanied by a significant decrease in the F: M load.

Hoilijoki and coworkers (1999) performed a bench-scale study observing effects of low temperature on nitrification in activated sludge (with and without carrier plastic) treating anaerobically pretreated municipal landfill leachate. The temperature regime consisted of 149 days at 10°C then 21 days at 7°C and finally 16 days at 5°C. At 10°C, complete ammonium removal was obtained in both reactors with volumetric loading rate of 0.05 g NH<sub>4</sub><sup>+</sup>-N l<sup>-1</sup> d<sup>-1</sup>, ammonium loading rate up to 0.02 g NH<sub>4</sub><sup>+</sup>-N g MLVSS<sup>-1</sup> d<sup>-1</sup> and HRT down to 2.7 days. The decrease of temperature to 7°C was accompanied by a slight increase in effluent NH<sub>4</sub><sup>+</sup>-N concentration. However, over 93% ammonium removal was obtained at 7°C with similar loading rates and HRT as at 10°C. Ammonium removal in the activated sludge reactor was only partial at 5°C with a removal rate of 61% (effluent NH<sub>4</sub>-N concentration 10–35 mg l<sup>-1</sup>). Hoilijoki suggested that this was due to change in sludge quality at the end of the study period, a conclusion arrived at through visual observation.

## **2.6 pH and Alkalinity**

Biological processes are effected by the pH of the surrounding environment. In wastewater treatment, biological mediated processes are subject to a wide range of pH fluctuation. Reactor pH conditions have been found to have a significant effect on the rate of nitrification (USEPA, 1993). Early work by Downing *et al.* (1964) measured nitrifying activity (as opposed to growth rate) in activated sludge as a function of pH. Those researchers showed that activity was fairly flat topped between pH values 7.5 – 8.5 with little or no activity below pH 6.5 and above pH 10. Painter and Loveless (1983)

reported pH optimum between 7.5 – 8.0, although in one experiment at 16°C, the highest growth rate was achieved at pH 8.5. Also at pH 6.5 the growth rate constant was between one third and two thirds of the maximum with no nitrification occurring below pH 6.0. Groeneweg *et al.* (1994) reported similar pH effects on ammonia oxidation at ammonia concentrations near  $K_s$ . Maximum rate of ammonia oxidation was obtained between pH 7.5 – 8.0 with the rate being fairly consistent between pH of 6.7 and 9.0. Between pH 6.0 – 6.7 the activity dropped sharply while some activity was recorded at pH 11.0. Groeneweg *et al.* (1994) noted similar results to previous research by Prakasam and Loehr (1972) who showed that ammonia oxidation is possible up to pH 11.2. The USEPA (1993) cites research by Poduska and Andrews (1975) who show a significant deterioration of nitrification from a sudden pH shift from 7.2 to 5.8. Poduska and Andrews (1975) observed rapid improvement in ammonia oxidation when pH was returned to 7.2, resulting in the conclusion that the effect of pH is only inhibitory and non-toxic. Watson *et al.* (1989) reported pH range for growth of pure cultures of ammonia oxidizers is 5.8 to 8.5 also noting that nitrification was inhibited below pH 5.8 from preliminary experiments performed with an enriched culture of nitrifiers obtained from wastewater. Prinic *et al.* (1998) cites work from DeBoer *et al.* (1989) and Walker and Wickramasinghe (1979) who show that in natural environments, such as soil, nitrification has been observed at pH below 4.0. This is supported by other studies that show where wastewater treatment systems have acclimated to low pH producing successfully nitrified effluent (Hoilijoki, 2000).

Even though the growth rate for nitrifying systems is effected by pH, research on the effect of pH on the saturation constant,  $K_s$ , showed relatively consistent values over a

typical pH range for wastewater treatment. Neufeld *et al.* (1980), referenced by Groeneweg *et al.* (1994), obtained a constant  $K_S$  for free  $\text{NH}_3$  of 0.152 mg/L  $\text{NH}_3$  over a pH range from 7 to 9. Jones and Morita (1985) also demonstrated the consistency of  $K_S$  over a practical range of pH. Jones and Morita (1985) grew a marine *Nitrosomonas* sp. at 5°C at varying pH values reporting a  $K_S$  for free  $\text{NH}_3$  decreased slightly from 0.014 mg/L  $\text{NH}_3\text{-N}$  at pH 7.8 to 0.011 mg/L  $\text{NH}_3\text{-N}$  at pH 6.8. Conversely previous work by Drozd (1976) reported a decrease of  $K_S$  for free  $\text{NH}_3$  from 0.22 mg/L  $\text{NH}_3$  at pH 9 to 0.07 mg/L as  $\text{NH}_3$  at pH 6 for a batch culture of *N. europaea*. Laudelout *et al.* (1976) reported a similar trend where  $K_S$  for  $\text{NH}_3$  was 1.15 mg/L  $\text{NH}_3\text{-N}$  at pH 8 and 0.04 mg/L  $\text{NH}_3\text{-N}$  at pH 6.

When discussing nitrification and pH, alkalinity is an important factor to consider. Nitrification is carried out by chemoautotrophic bacteria, which utilize inorganic carbon for growth. The majority of inorganic carbon in water exists as dissolved carbon dioxide,  $\text{H}_2\text{CO}_3$  (carbonic acid), and the subsequent deprotonated species,  $\text{HCO}_3^-$  (bicarbonate) and  $\text{CO}_3^{2-}$  (carbonate). The sums of these carbonate species that can accept a proton ( $\text{HCO}_3^-$  and  $\text{CO}_3^{2-}$ ) are defined as alkalinity. Alkalinity is a typical measure of the buffering capacity of water measured in mg/L of  $\text{CaCO}_3$  (calcium carbonate). It can be shown that over a pH range of approximately 5 to 8 in an aqueous biological reactor, the equilibrium pH of the reactor will be dictated by the amount of alkalinity and  $\text{CO}_2$  present in the system (USEPA, 1993). Recall from Equation 8 that stoichiometrically the theoretical alkalinity consumption is 7.1 mg (as  $\text{CaCO}_3$ ) per mg of ammonia-nitrogen oxidized. The majority of alkalinity consumed is due to acidification caused by oxidation of ammonia to nitrate. There is also uptake of aqueous carbon dioxide for autotrophic biomass



synthesis; however the amount of alkalinity consumed is relatively small due to slow nitrifier growth rates. Alkalinity can also be lost through stripping in aerated systems and should be accounted for in full-scale system design (USEPA, 1993).

## 2.7 Dissolved Oxygen

It has been shown previously that the total oxygen requirements in a biological system are related to the oxygen consumed to supply energy for synthesis and the oxygen consumed for endogenous respiration (Eckenfelder, 1970). This assumes that oxygen must be supplied to the system in order to satisfy the following demands: (1) biological organic removal, (2) endogenous respiration where cells lyse and release soluble oxidizable organic compounds, (3), chemical oxidation as measure by the immediate oxygen demand, and (4) ammonia which is oxidized to nitrate (Adams, 1978). Based on stoichiometry (Equation 10), the theoretical oxygen utilization for ammonia oxidation is 4.57 mg O<sub>2</sub> required per mg ammonia-nitrogen oxidized (USEPA, 1993). However, these observations have been questioned (Sharma, 1976; Churchwell, 1980), on the basis that a small amount of oxygen is produced in the process of protoplasm synthesis. Investigation by Wezernak and Gannon (1967) based on 120 bottle BOD studies, concluded that 3.22 mg O<sub>2</sub> is required per mg NH<sub>3</sub>-N oxidized to NO<sub>2</sub><sup>-</sup>-N, and that 1.11 mg O<sub>2</sub> is required per mg NO<sub>2</sub><sup>-</sup>-N oxidized to NO<sub>3</sub><sup>-</sup>-N. Based on this study, the total observed oxygen demand for oxidation of NH<sub>3</sub>-N to NO<sub>3</sub><sup>-</sup>-N is 4.33 mg O<sub>2</sub> per mg NH<sub>3</sub>-N. The concentration of DO has a significant effect on the rates of nitrifier growth and nitrification in biological waste treatment systems (USEPA, 1993). Using the Monod equation to model growth of *Nitrosomonas* with DO as the growth limiting substrate concentration, values for the half-saturation coefficient have been reported as 0.15 – 2.0

mg/L O<sub>2</sub> (USEPA, 1993). Stenstrom and Song (1991) studied oxygen limitation on the nitrification process in activated sludge concluding that the values of DO at which nitrification is limited can be between 0.5 to 2.5 mg/L in either suspended or attached growth systems under steady state conditions. Also a high solids retention time was suggested to ensure nitrification at low DO levels. Furthermore Stenstrom and Song (1991) reported that under transient conditions of organic shock loading, diffusional resistance and heterotrophic/nitrifier competition can increase the limiting DO value significantly. Hanaki *et al.* (1990) also reported similar findings. As suggested by Ekama *et al.* (1984), the wide range of values for the saturation coefficient with respect to DO probably has arisen because the concentration of DO in the bulk liquid is not necessarily the same as inside the biological flow where the oxygen consumption takes place. Consequently the value will depend on the size of floc, mixing intensity and oxygen diffusion rate into the floc. For these reasons it is not really possible to establish a generally applicable minimum oxygen value (Ekama *et al.*, 1984). Temperature, SRT, pH and DO are important parameters in optimizing the activated sludge process. This study will investigate temperature and SRT effects on bench-scale operation using municipal wastewater from the community in an attempt to simulate treatment performance at the local full-scale WWTP.

## **Chapter 3.0**

### **Materials and Methods**

#### **3.1 System Specifications**

The treatment system used during this bench-scale study was modeled after the Kuwahee wastewater treatment activated sludge system. This facility incorporates six nitrification activated sludge reactors, with a total volume of 31040 cubic meters, in the treatment process. Following aeration, the mixed-liquor flows by gravity into five 41.2 meter diameter circular final clarifiers. The settled nitrifying sludge is returned to the aeration tanks via a biosolids wet well. The bench-scale treatment system has been designed to simulate overall operation in the aeration basins and secondary clarifiers of the full-scale facility.

#### **3.2 Reactor Vessel**

Figure 3 depicts the laboratory scale activated sludge system. The reactor/clarifier combination was one of four systems used during the study. The utilization of four reactors allowed for simultaneous operation at differing SRTs. Figure 4 shows all four reactors which comprise the entire treatment system. Each reactor vessel was constructed from 25.4 cm (10") diameter acrylic tubing with 0.635 cm (1/4") thick sidewalls for the mid-section and 1.27 cm (1/2") acrylic plating for the top and bottom. The bottom plate was routed so the cylindrical midsection fit snugly to ensure proper adhesion of the two pieces and also to prevent leakage of the reactor contents. The bonding of the midsection to the bottom plate was accomplished using an acrylic adhesive and a proper seal was insured through the application of silicone caulk on the

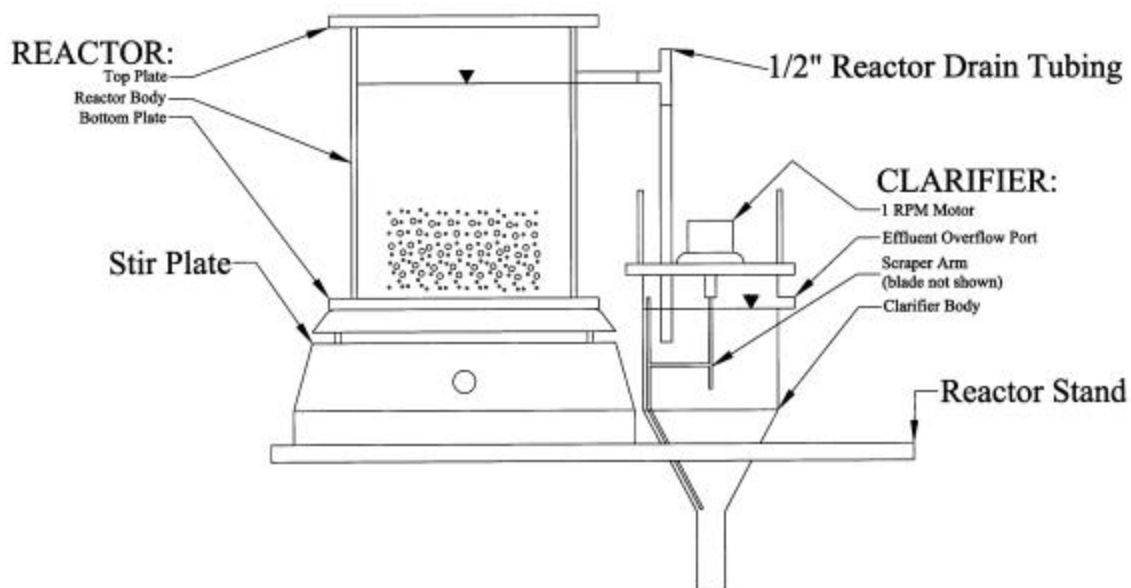


Figure 3. Schematic landscape illustration of lab scale unit.



Figure 4. A picture of the entire treatment system consisting of the reactors and clarifiers.

interior of the vessel at the seam between the midsection and the bottom plate. The top plate was also routed to provide a proper fit and was secured to the reactor vessel by four kurneled posts and wing nuts running from the bottom to the top plate. The top plate was ported to allow the insertion of various probes and to prevent pressurization of the interior headspace due to aeration. The reactor was ported for gravity overflow of the contents to the external clarifier. The elevation along the cylindrical sidewall of the reactor vessel for the gravity overflow port was chosen to provide a 10-liter operational volume for the vessel. A clear polypropylene fitting with 1.27 cm (½") NPT tread and 1.27 cm (½") hose barb was installed into the treaded port. To monitor clogging of the exit port, a clear fitting was used for ease in visual inspection of solids build-up. The gravity overflow allowed the mixed-liquor to flow into a 1.27 cm (½") T-connector, providing atmospheric pressure equalization, before exiting the system via 1.27 cm (½") clear tubing vertically downward to the external clarifier.

### **3.3 Mixing System**

The activated sludge process is a suspended-growth culture of microorganisms existing within a continuous flow regime. The mixing of the 10-L reactor vessel was achieved using a magnetic stir plate. The use of a mechanical means to mix the reactor contents was deemed necessary due to the insufficient amount of mixing provided through aeration at low air flow regimes. A 38.1 cm (15") square Thermolyne™ magnetic stirrer was used to provide adequate mixing and a stable base to support the reactor. A 7.6 cm (3 in.) long, 1.27 cm (½") diameter Teflon®-coated stir bar was used as the impeller. The mixing intensity was gauged visually through the movement and distribution of the flocs within the reactor. The dial, which controls the speed of the stir

bar, was set at approximately  $\frac{1}{2}$  speed. The stir plates were typically set to approximately one-half their maximum setting of 1000rpm, which corresponds to a power output of 15 watts. Parker (2002) presented a calculation which showed that the reactors utilized during this study were completely mixed.

### **3.4 External Clarifier with Scraper Arm**

The external clarifier was a necessary part of the bench-scale treatment system in order to simulate biosolids recycle at the full-scale municipal WWTP. The full-scale facility utilizes five final clarifiers with dimensions of 41.2 meters in diameter and 3.7 meter water depth, which operate in parallel. With an average daily flow of 151416 m<sup>3</sup>/d, the HRT for each full-scale final clarifier is approximately 3.85 hours. However, for the bench-scale system, a fixed flow rate of 19 mL/min was used in order to meet the hydraulic parameters for the reactor vessel. At this fixed flow rate, the required volume of the final clarifier for the bench-scale system would be 4.6 L to meet the 3.85-hour retention time at the full-scale plant. To keep the clarifier volume reasonable, a lower HRT (2.2 hr) was chosen. This HRT was thought to be sufficient to provide adequate time for biomass settling. The flow rate and HRT established the working capacity of the clarifier at 2.5 liters. Lab Glass, a commercial glass blowing company, constructed each external clarifier per the following design. The upper cylindrical midsection was 15.2 cm (6 in.) in diameter with a 0.952 cm ( $\frac{3}{8}$  in.) O.D. port to provide gravity overflow of the clarified effluent. The placement of this port fixed the operational volume of the clarifier at 2.5 L. The conical midsection was specified to be 53° in pitch to the horizontal. The final cylindrical section was to be 3.8 cm (1  $\frac{1}{2}$  in.) long and 0.952 cm ( $\frac{3}{8}$  in.) O.D. to adapt to the tubing which would recycle the settled biosolids back to the reactor.

The other component of the clarifier was a scraper arm which was used to facilitate settling of the MLSS and to prevent adhesion and growth of bacteria on the sidewalls of the clarifier. This scraper arm assembly consisted of several parts; a scraper arm, a small motor and an acrylic housing for these components. The scraper arm was composed of a stainless steel wire approximately 3 mm (0.118 in.) in diameter fitted with a Triple-Edge™ wiper blade. The wiper blade had a hollow annular space permitting the wire to be inserted. The wiper blade was necessary to provide good contact between the scraper arm and the interior of the glass clarifier. This scraper arm was attached to a fitting using a small setscrew and fixed to the drive shaft of a 3-rpm electric motor. The motor was mounted to 0.635 cm (¼ in.) thick acrylic plate which measured 17.8 cm (7 in.) square with a routed circular groove to provide a secure fit to the top of the clarifier. A 15.2 cm (6 in.) cylindrical acrylic upper portion was bonded to the motor mount plate. This provided partial protection from any electrical hazards and protected the cooling fan on the motor from any contact that might cause damage.

After this design was employed, several changes (including the use of a slower motor and a timer) were made to the clarifier assembly. The 3-rpm motor operating continuously provided excess mixing in the clarifier, thus hindering MLSS settling. Therefore, a 1-rpm motor was chosen to reduce the speed of the scraper arm as it swept the clarifier walls. In order to impart as little mixing in the clarifier as possible, a timer was installed and set to turn the motor on for 30 seconds at 5 minute intervals. These changes were sufficient to prevent adhesion of solids to the walls of the clarifier. The other changes made to the clarifier were in the alteration of the constructed glass vessel. The conical midsection of the clarifier was drawn down from 53° to approximately 70°

pitch to the horizontal. This provided a greater slope to the clarifier bottom that aided in the collection of solids needed to provide efficient recycle. The 0.952 cm ( $\frac{3}{8}$  in.) cylindrical section following the conical section was replaced with a 7.6 cm (3 in) long and 3.18 cm ( $1\frac{1}{4}$  in.) diameter cylindrical section of glass tubing. This piece was welded onto the newly drawn conical midsection creating a reservoir for the collection of the solids from the reactor overflow. The addition of this piece and the newly drawn conical section increased the clarifier volume from approximately 2.5 L to 2.7 L which subsequently increased the HRT of the clarifier from 2.2 hours to 2.4 hours. All changes described were applied to each individual bench-scale system before any treatment performance data were collected.

### **3.5 Influent Feed/Recycle System**

Influent wastewater was collected after primary clarification from the Kuwahee WWTP and stored in a 378.5 L (100 gal.) reservoir. Wastewater was drawn from the reservoir tank using a common 0.952 cm ( $\frac{3}{8}$  in.) i.d. Tygon tube feed line connected to a digitally controlled Cole-Parmer© peristaltic pump. The pump had four peristaltic pump heads attached to the main drive. The common feed-line was split into four separate feed lines (one for each reactor). These individual feed lines were also 0.952 cm ( $\frac{3}{8}$  in.) i.d.. Tygon tubing similar to the common feed line. The split flow resulted in a flow rate of 19 mL/min into each reactor vessel. Each individual feed line was attached to a 90° elbow fitting screwed into the upper portion of the posterior side of the reactor vessel. This wastewater feed then passed through an identical fitting on the interior of the reactor which allowed for the wastewater feed to drip into the reactor. Without this second 90° elbow, the wastewater feed would run down the interior sidewall into the reactor. To



prevent hydraulic short-circuiting, it was necessary to introduce the feed more toward the center of the reactor.

The recycle flow regime was similar to the influent feed system. The recycle line leading from the bottom of the clarifier passed through a peristaltic pump identical to that described for the influent feed system. However, each individual recycle line passed through a peristaltic pump head and was directly connected to the reactor feeding that specific clarifier. Therefore, solids collected from each bench-scale system were pumped directly back into that system. The recycle flow rate was set at 19 mL/min providing a recycle ratio of 1.

### **3.6 Aeration System**

Since the activated sludge process is aerobic, the supply of air is an integral component of the system. During this work, four activated sludge reactors were operated at an excess level of dissolved oxygen with respect to nitrification ( $\text{D.O.} \geq 2.0 \text{ mg/L}$ ). Therefore, an efficient and constant supply of oxygen to the treatment system was needed. A compressed air supply, available at the research facility, was used as the oxygen source. Since the pressure provided in the house air line is approximately 70 psi, a section of reinforced tubing was needed to connect the house air to a regulator to step down the pressure. After passing through a pressure regulator, the house air was then humidified before being fed into the reactors. Air humidification was accomplished by using a 20 L polypropylene carboy containing approximately 5 L of water with the air supply directed through a stainless steel tube that was submerged in the water. After passing through the water column in the carboy, the air exited through the top of the carboy. The assembly at the top of the carboy consisted of an air-tight rubber stopper

which fit inside of a screw cap with an opening for two stainless steel tubes; one submerged in the water column (inlet) and one that was not submerged (outlet). After the air exited the humidification chamber, it was directed through a flow manifold with a common inlet and four separate outlets. The common inlet fed the air through four separate flow tubes, each of which was dedicated to an individual reactor. Each flow tube gave a direct readout of the airflow passing through each individual tube (in  $\text{cm}^3/\text{min}$ ). This corresponded to the flow of air entering the reactors. Small aluminum valve cartridges provided adjustment of the air flow rate and a spherical glass float in the flow tubes provided a direct read measurement of the quantity of air entering the reactors. A dead leg was also installed before the air entered the flow manifold to collect any moisture that would condense on the inside of the tubing that supplied the air to the flow manifold. The dead leg was clamped during normal operation; however, the clamp was released daily to blow out any moisture that collected during that days operation. After the air had been regulated, humidified, and flow controlled, it was split and fed into each reactor via two air lines coupled to 15.2 cm (6 in) long cylindrical aquarium style air diffusers. These air diffusers were chosen for the fine bubbles they produced; however, the nominal pore size was not defined by the vendor for these tubes making it difficult to determine mass transfer levels. However, the flow manifold in combination with the air diffusers allowed for the coarse control of the dissolved oxygen concentration in the reactors. This level of control was sufficient to maintain excess levels of dissolved oxygen ( $\geq 2.0 \text{ mg/L}$ ) in the mixed-liquor.

### **3.7 Light Attenuation**

The attenuation of light was important so as not to facilitate the proliferation of phototropic organisms present in the activated sludge, such as algae. The reactors were operated in a windowless darkroom although fluorescent lighting was available. These lights remained off except for periods of sampling or reactor maintenance.

### **3.8 System Start Up**

#### **3.8.1 Reactor Seeding**

The activated sludge added to the aerated, continuous flow, reactors was collected from the Kuwahee Municipal WWTP. A modified version of the Monod equation was used to estimate start-up MLSS concentrations. Using “typical” values for municipal wastewater treatment, the calculated MLSS concentrations for the 20, 10, 5 and 2 day SRT reactors were 2328, 1552, 931 and 423 mg/L. An ample amount of mixed-liquor was collected from the waste activated sludge feed to the final clarifier at the Kuwahee Municipal WWTP. All reactors required the dilution of the collected mixed-liquor except for the 20-day SRT reactor which required concentrating the mixed-liquor to achieve the appropriate concentration. Concentration the mixed-liquor was achieved by allowing the collected sludge was allowed to settle before a specific volume of clear water was decanted from the vessel. Supernatant removal resulted in a higher concentration of MLSS. The four reactors were seeded on September 28, 1999.

## 3.9 System Operation

### 3.9.1 SRT Control

Four bench-scale systems were operated in parallel at different SRTs (20, 10, 5 and 2-days) over the duration of this study. The upper end of this range (20 and 10 day SRT) was more representative of the SRT used at the full-scale WWTP, whereas the 2- and 5-day SRTs were included to stress nitrification performance. Solids retention time relates the quantity of microbial solids in an activated sludge process to the rate of biomass lost in the effluent and withdrawn in the waste sludge (Viessman and Hammer 1993). The equation used to calculate SRT is presented here as Equation 18.

$$q_c = \frac{VX}{Q_w X + (Q - Q_w) X_e} \quad (18)$$

Various methods of controlling the SRT in each of the reactors had been considered. Due to the small volumes utilized in the bench-scale system, a pump could not be employed to remove MLSS at a specified wastage rate as traditionally done in many full-scale facilities. Instead, MLSS were wasted directly from the aeration tank on a daily basis. Various wastage methods were tested to control SRT including wasting an average amount from the reactors daily, wasting a volume of the reactor contents based on the previous days solids analysis, and daily wastage from all four reactors based on a solids analysis for that day. The method that provided the highest level of control was daily wastage from all four reactors based on the solids analysis for that day. Solids analysis is discussed in further detail later in this chapter. Having determined MLSS and

effluent suspended solids (ESS) for that day, the wastage flow rate,  $Q_w$  was determined. The 2-day reactor required wastage volumes almost equal to one half of the total volume of the reactor. To minimize the probability of removing the microorganisms at a rate faster than they can reproduce, half of the wastage volume was removed during the morning and the other half removed during the afternoon for the 2-day reactor.

### **3.9.2 Temperature Control**

Since temperature was a key variable in this work, proper temperature control was important. The reactors were operated at 3 different temperatures during this study, 22, 12 and  $7^{\circ}\text{C}$ . To provide appropriate temperature control, the reactors were operated in a constant temperature room with a digitally controlled HVAC unit to maintain a set temperature within the room.

### **3.9.3 Dissolved Oxygen Control**

The dissolved oxygen concentration was controlled using an air flow meter. As discussed in the section describing the aeration system, the air introduced into the reactors was controlled by individual air flow meters which provided a direct readout of the air flow rate into the reactors. A DO probe was used to periodically check the dissolved oxygen levels in the reactors and, if necessary, adjustments in the air flow rate were made to maintain an adequate DO concentration in the reactors. The DO concentration was maintained above 2.0 mg/L to minimize adverse effects on nitrification.

### **3.9.4 Waste Collection/Preservation**

Influent municipal wastewater was collected from the Kuwahee WWTP approximately every four days. Two 208.2 liter (55 gallon) drums were filled with wastewater (after primary clarification) using a ½ horsepower sump pump. The drums were then transported to the laboratory and stored at 5°C. In order to maintain the continuity and homogeneity of the wastewater, the contents of the drums were pumped into a 378.5 liter (100 gallon) HDPE closed dome tank and mixed. Initially mixing of the wastewater was accomplished using periodic operation of a 1/3 horsepower mixer with a shaft driven impeller. This mixer was mounted on a custom made cap fitted with a 5.1 cm (2") NPT bung hole. The cap was attached at the top of the tank so that the shaft of the mixer would extend vertically downward into the contents of the 378.5 liter tank. However, the mixing intensity using this mixer was too great to provide moderate recirculation of the wastewater during storage. The mixing was so intense that headspace oxygen was imparted to the wastewater and some degradation of BOD occurred during storage. In order to homogenize the tank contents without aerating, a small electric re-circulation pump was used. However, the pump had a horizontal intake that directed the wastewater vertically upward causing problems when the volume of influent wastewater in the holding tank was low. Therefore, the re-circulation pump was fitted with a T-connector on the outlet so the flow leaving the pump would not breach the surface of the wastewater in the tank. This eliminated the possibility of aeration of the wastewater before being fed to the reactors, and also adequately blended the holding tank contents.

### **3.9.5 Sample Collection**

Sample collection was performed every one to two days. The types of analysis performed to record reactor performance are presented in the following sections of this chapter. In general, the performance of the bench-scale system during operation was contingent on several variables. The documented data for each reactor include solids, nitrogen, alkalinity, and chemical oxygen demand analysis. There was a limit to the number and frequency of tests performed due to costs and available resources. Thus, there are some diagnostic parameters which were not analyzed. However, an adequate amount of data was recorded to present a fairly comprehensive study on temperature effects on nitrification in activated sludge.

## **3.10 Sampling and Analysis Procedures**

### **3.10.1 Solids Analysis**

#### *Mixed Liquor Suspended Solids (MLSS)*

Daily analysis of the MLSS concentration was required for the maintenance of SRTs for each of the four systems. The MLSS samples were analyzed according to Standard Method 2540-D (Total Suspended Solids Dried at 103-105°C) (APHA, 1998). The procedure for the collection of samples was specific for the bench-scale system constructed for this research. Initially, the effluent drainage port on the reactors was plugged while the influent pump was turned off. The recycle pump flow rate was increased until all the solids from the secondary clarifiers were pumped into the reactors. This step was performed to ensure that all the solids present in the entire system were in the reactor vessels during sampling. The recycle pump, aeration system, and stir plates

were then turned off and the solids in the reactor vessel were allowed to settle somewhat. After the solids settled, the reactors were unplugged and the clear supernatant was allowed to decant into the clarifiers. The aeration and stir plates were turned back on to resuspend the solids in each reactor; however, both the feed and recycle pumps remained off. Once the solids were resuspended, the mixed-liquor sample was then collected. A predetermined sample volume of mixed-liquor was removed from each reactor to provide an accurate measurement of MLSS concentrations.

#### *Mixed Liquor Volatile Suspended Solids (MLVSS)*

In an effort to better quantify the microbial fraction of the MLSS in each reactor, Standard Method 2540-E (Fixed and Volatile Solids Ignited at 550°C) was conducted daily (APHA, 1998). Special filters prepared for the determination of VSS were used during the determination of MLSS. Filters containing dried suspended solids were placed in a small muffle furnace and ignited (550°C) for a minimum of 15 minutes.

#### *Effluent Suspended Solids (ESS)*

The effluent suspended solids, unlike the MLSS, were determined through the use of a 24-hour composite sample. Effluent obtained over 24 hours of operation was collected in a 20 L polypropylene carboy. The carboy containers were stirred to homogenize the composite sample before pumping approximately 300 to 400 mL of effluent into an Erlenmeyer flask. Samples were then filtered through 0.45-micron pre-weighed filters. Care was taken to wash any solids that may have adhered to the filtration flask onto the filter. These samples were analyzed according to Standard Method 2540-D (Total and Suspended Solids Dried at 103-105°C).



### 3.10.2 Nitrogen Analysis

The speciation dynamics of nitrogen during operation of the bench-scale treatment systems was integral in the evaluation of environmental changes on nitrification treatment performance. For this reason, several species of nitrogen were analyzed over the course of this study.

#### *Ammonia ( $\text{NH}_4^+$ )*

Ammonia, the substrate compound for ammonia oxidizing bacteria, was measured on a daily basis for each reactor system. Ammonia samples were taken from the effluent ports on the external clarifiers for each treatment system. The influent ammonia sample was drawn directly from the 378.5 L holding tank. A 100 mL sample was collected in a small 150-mL Erlenmeyer flask, which also served as the container in which the analysis was performed. These flasks were stoppered and allowed to come to room temperature (approximately 30 minutes) before analysis using an ion selective electrode (Orion Model # 95-12). Before each analysis the probe was calibrated to a set of freshly prepared standards (50, 5, 0.5, and 0.1 ppm  $\text{NH}_4^+$  as N). Additionally, a quality control sample was prepared. Calibration for the ammonia probe was a point to point fit to minimize any error due to the non-linearity of the response over the range of ammonia concentrations used during the calibration. The probe readings were documented on a data analysis sheet. The analysis for ammonia in water and wastewater is referenced as Standard Method 4500D, Ammonia Selective Electrode Method (APHA, 1998).

### *Anion Sampling and Analysis*

The biological transformation of ammonia produces distinct nitrogen compounds. The first is nitrite ( $\text{NO}_2^-$ ) which is an intermediate compound before the second transformation to the nitrate ( $\text{NO}_3^-$ ) end product. Measurement of nitrite and nitrate was determined using ion chromatography. The grab sample taken in the 150 mL Erlenmeyer flask was used to draw 5 mL of sample into a sterile 10-mL lure-lok™ syringe. A small Acro-disk™ 0.45  $\mu\text{m}$  syringe filter was then attached to the syringe and the sample filtered into labeled 5-mL vials, fitted with special caps, for processing using the auto-sampler on the ion chromatograph.

Processing the anion samples falls within certain EPA guidelines for preservation, if the samples are not to be run shortly after collection. For nitrite and nitrate the preservation technique is to simply cool the aqueous sample to 4°C and store for no longer than 48 hours before analysis. This protocol was followed during operation of the ion chromatograph (IC). However, technical difficulties were experienced with the IC during this study. During this down time samples were stored at 4°C, but for periods up to 28 days. After the instrument was repaired, a few anion samples were analyzed both immediately after collection and at storage times up to 28 days. Little change in sample concentration was measured over time.

The anions  $\text{NO}_2^-$  and  $\text{NO}_3^-$  were analyzed using ion chromatography. Standards were prepared within a range encompassing the expected concentration commensurate for each analyte. These ranges consisted of 0.1 mg/L to 40 mg/L as N for nitrate and 0.1 mg/L to 20 mg/L as N for nitrite.

Standard Method 4110.B references the method of analysis of these anions, Ion Chromatography with Chemical Suppression of Eluent Conductivity (APHA, 1998). The analysis was performed using a Dionex<sup>®</sup> DX500 Ion Chromatogram with an AS40 Dionex Auto-sampler. The minimum detectable concentration of an anion is a function of sample size and conductivity scale used. According to Standard Method 4110 B, minimum detectable concentrations are near 0.1 mg/L for  $\text{NO}_3^-$  and  $\text{NO}_2^-$  with a 100  $\mu\text{L}$  sample loop and a 10  $\mu\text{S}/\text{cm}$  setting on the conductivity detector. The output data from the IC was in the form of area for individual peaks, which represent the change in conductivity during the elution of the specific anion. The area for the analyte was used to determine concentration through calibration of the IC.

### **3.10.3 Sludge Volume Index (SVI)**

SVI typically is used to monitor settling characteristics of activated sludge and other biological suspensions. The SVI was determined using Standard Method 2710 D, Sludge Volume Index (APHA, 1998). One liter of MLSS was pumped, using a peristaltic pump, from a reactor into a 1 L graduated cylinder. The contents of the graduated cylinder were completely mixed to ensure an initially homogeneous mixture. The settled volume of biosolids was assessed after 30 minutes. Using the settled volume and the MLSS concentration, the SVI was obtained.

### **3.10.4 Chemical Oxygen Demand ( $\text{COD}_T$ )**

Chemical Oxygen Demand is the oxygen demand exerted on a wastewater by oxidizable organic matter. The  $\text{COD}_T$  test employed during this research measured the total amount of oxidizable organic matter in a sample. The quantification of treatment

efficiency with respect to COD<sub>T</sub> removal was performed by analysis of common influent grab samples and composite effluent from each of the four reactor systems. The influent and effluent samples collected did not undergo filtration to remove suspended colloidal or inert solids, thus the subscript 'T' in COD<sub>T</sub> is used for this reason. The samples were preserved in concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) at a ratio of 2-mL H<sub>2</sub>SO<sub>4</sub> per 100-mL sample. This preservation technique is accepted by the USEPA and suggested by Standard Methods (APHA, 1998). The samples were collected in sterile 15 mL centrifuge tubes and preserved with 30 µL of sulfuric acid. After acidification, the samples were stored at 4°C until analysis (<28 days).

Analysis of COD<sub>T</sub> was performed as stated in Standard Method 5220 D, Closed Reflux Colormetric Method (APHA, 1998). The COD vials used during the experiment consisted of two ranges, low range: 0-150 mg COD and high range: 0-1500 mg COD. Effluent samples were generally analyzed using the low range vials and influent samples were analyzed using the high range vials. 2 mL of sample was added to the Bioscience™ vials and 2.5-mL sample was added to the HACH® vials. Standards were prepared using potassium hydrogen phthalate. A 1000 mg COD per liter stock solution was prepared, and subsequent dilutions were drawn to generate a range of standards commensurate with the analysis range of the COD vials. A set of four standards and one blank (0.2 µm filtered, DI water) were analyzed with the samples. A Perkin-Elmer Spectrophotometer Model # 3B was used to measure the absorbance of light at 440 nanometers (nm) for the low range vials and at 680 nm for the high range vials. Disposable methylacrylate cuvettes were used for colormetric readings.

### **3.10.5 Alkalinity**

Alkalinity provides pH buffering capacity to neutralize acidifying processes such as nitrification. Therefore, the alkalinity present in the untreated wastewater must be sufficient for nitrification to proceed uninhibited. Alkalinity measurements were performed using Standard Method 2320 B., Titration Method (APHA, 1998). The influent and the four effluent samples were monitored for alkalinity. Samples were taken with varying frequency, usually once to twice a week and in concurrence with the ammonia test. Due to the volume sensitive nature of the test, 100 mL volumetric pipettes were used to draw exactly 100 mL of sample and dispense into a beaker where the titration was performed.

### **3.10.6 pH/Temperature**

The pH and temperature of each reactor were monitored discretely to ensure proper operation. An Orion model pH probe with automatic temperature correction was used for the pH measurements. This probe model also provided a display of temperature of the reactor contents. Reactor operation was performed in a constant temperature room which provided a constant air temperature environment; however, mixed liquor temperature was monitored for any variation from the room set points.

### **3.10.7 Dissolved Oxygen (DO)**

Dissolved oxygen is a measure of oxygen activity in the aqueous environment and provides a measure of oxygen available to microorganisms. The activated sludge process is aerobic and therefore requires a sufficient amount of oxygen to proceed uninhibited. In this study, the four bench-scale systems were operated at excess DO concentrations.

Previous research has indicated that at DO levels above 2.0 mg/L, inhibition of nitrification does not occur. Therefore, the DO in each reactor was maintained above 2.0 mg/L.

## Chapter 4.0

### Results and Discussion

#### 4.1 COD Treatment Performance

To ensure that the temperature remained constant, the reactors were placed in a constant temperature room for the duration of the study. The set points for this study were originally 20, 10 and 5°C; however, the temperature maintained within the constant temperature room remained a degree or so higher than the set point. Actual wastewater temperatures in each reactor over time are presented in Table 2. During operation at 20°C, the average reactor temperature was 21.7°C with a standard deviation of 0.15°C. For operation at 5°C, the average reactor temperature was 6.6°C with a standard deviation of 0.2°C and for operation at 10°C, the average reactor temperature was 11.5°C with a standard deviation of 0.14°C. Despite the disparity between the set point and actual temperature maintained within the reactors, the constant temperature room allowed for tight control of mixed-liquor temperature during the experiment.

Table 2. Average wastewater temperatures measured during this treatability study.

Reactor SRT	Phase I 20°C Set Point	Phase III 10°C Set Point	Phase II 5°C Set Point
20	21.68	11.61	6.76
10	21.65	11.50	6.65
5	21.66	11.52	6.67
2	21.61	11.55	6.48

The study of temperature and SRT effects on activated sludge treatment performance of municipal wastewater was begun on September 28, 1999 (day 1). During the course of this study, the 20, 10, 5 and 2 day SRT reactors were operated at 22, 7 and 12°C for 61 days (September 28, 1999 to November 27, 1999), 46 days (November 28, 1999 to January 12, 2000) and 105 days (January 13, 2000 to April 27, 2000), respectively. The reactors were initially operated at the highest temperature utilized in this study (22°C) for a period of 61 days. The low temperature (7°C) portion of this study was begun on day 62 and concluded on day 107 when it was determined that steady state nitrification was not being achieved. The temperature was then raised to 12°C on day 108 for the remainder of the study to reestablish steady state nitrification in all four reactors.

Steady state for suspended growth reactors is an operational characteristic identified by equal rates of biomass growth and biomass loss, thus biomass accumulation is equal to zero (Benefield & Randall, 1985). Steady state is a balance between microbial growth from utilization of available substrate and biomass lost primarily from direct wastage and indirect loss of solids in the effluent. However, daily measurement of MLSS showed that steady state was affected by variations in wastewater composition and clarifier performance. This bench-scale study used actual municipal wastewater collected from the city WWTP rather than a synthetic wastewater, thereby providing a more applicable simulation of full-scale operation. Wastewater collected varied in strength as is consistent with day to day heterogeneity in composition. Figure 5 shows influent total COD ( $COD_T$ ) measured during the course of the study.



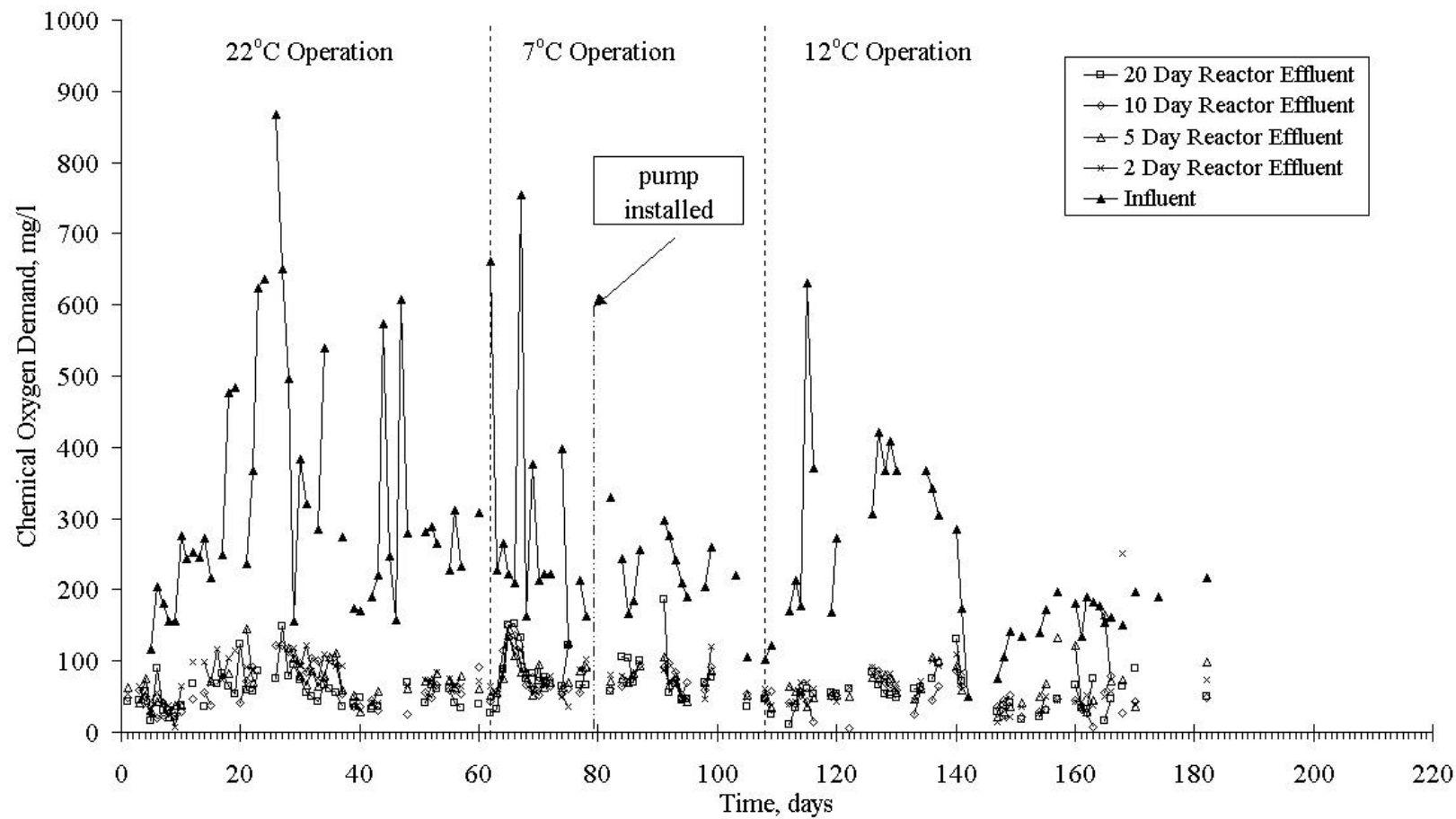


Figure 5. Effect of temperature (ranging from 22°C to 7°C) on COD<sub>r</sub> treatment performance.

Reactor operation initially proceeded for 22 days before a major perturbation in wastewater strength occurred. During operation of the bench-scale systems at 22°C, routine maintenance was performed at the Kuwahee WWTP on an anaerobic digester. The maintenance began on October 18<sup>th</sup> (day 21) and lasted until October 31<sup>st</sup> (day 34). During this time, wastewater and residual solids from the anaerobic digester were directed to the head of the WWTP for further treatment. A dramatic increase in wastewater strength resulted from this maintenance operation. Over the 13 day period, wastewater had approximately twice the concentration of ammonia and a similar change in COD<sub>T</sub>. The period during which the high strength wastewater was collected and fed to the reactors included the period from October 20, 1999 to November 2, 1999 (day 23 to day 36). The average COD<sub>T</sub> increased from  $249 \pm 43$  to  $496 \pm 131$  mg COD<sub>T</sub> / L (95% confidence interval). Data collected during this major perturbation were not used in steady state analysis.

Effluent COD<sub>T</sub> over time for each SRT is also presented in Figure 5. Effluent COD<sub>T</sub> measurements include the oxygen demand due to suspended solids since the effluent samples were not filtered during this treatment study. Table 3 summarizes the steady state COD<sub>T</sub> values and treatment efficiencies. During 22°C operation, effluent COD<sub>T</sub> decreased and treatment efficiency increased with increasing SRT. The values for carbon treatment efficiency were lower than typical values for municipal wastewater since COD<sub>T</sub> levels associated with effluent SS were included in measurements.

Mines and Sherrard (1999) also found that effluent COD decreased with increasing SRT while Bernal-Martinez et al. (2000) found that COD treatment efficiency increased with SRT for a reactor treating domestic wastewater.

Table 3. Summary of COD<sub>T</sub> data during defined steady state operation at various operational temperatures.

Reactor	COD <sub>T</sub> , mg/L		Treatment performance, %	
	Mean, (mg/L)	$\sigma$ , (mg/L)	Mean, %	$\sigma$ , %
<i>22 °C (day 37 to day 60)</i>				
Influent	282.8	125.4	–	–
20 day SRT	46.7	13.4	78.9	6.0
10 day SRT	50.6	17.3	77.2	7.8
5 day SRT	61.7	15.3	72.2	6.9
2 day SRT	62.7	19.3	71.7	8.7
<i>7 °C (day 72 to day 99)</i>				
Influent	262.2	139.6	–	–
20 day SRT	80.9	34.5	66.5	10.0
10 day SRT	72.7	15.6	67.3	7.0
5 day SRT	74.7	15.7	66.3	7.1
2 day SRT	74.1	22.3	66.6	10.0
<i>12 °C (day 116 to day 165)</i>				
Influent	212.8	116.6	–	–
20 day SRT	54.6	25.3	75.4	11.4
10 day SRT	46.9	22.9	78.9	10.3
5 day SRT	66.9	34.4	69.9	15.5
2 day SRT	58.1	25.5	73.8	11.5

Treatment efficiencies at 12°C did not follow the same pattern as treatment at 22°C; although, overall average steady state efficiencies were similar. The 10-day SRT at 12°C had the highest level of treatment followed by the 20, 2 and the 5-day SRT reactors. Using a two-sample t-test, the average treatment efficiencies for the 20 and 10-day SRT systems were not to be statistically different. Additionally, the 20 and 2-day SRT average treatment efficiencies were not statistically different at a confidence interval of 95%. However, the 5-day SRT average treatment efficiency was statistically different at a confidence interval of 95% from the other SRTs studied. No explanation could describe this observed effect between operational SRTs studied. During 7°C operation, treatment efficiencies were lower than those for the higher temperatures and were very similar at all SRTs studied. Using a two-sample t-test, the treatment efficiencies for all SRTs measured at 7°C were not statistically different at a confidence interval of 95%. A decline in COD<sub>T</sub> treatment efficiency with declining temperature was expected as reported in literature (Mines and Sherrard, 1999); however, this effect of temperature on carbon treatment efficiency was not apparent except during 7°C operation. This suggests that temperature rather than SRT governed carbon treatment for the given conditions.

In a temperature dependent study on COD treatment in activated sludge, Mines and Sherrard (1999) showed that effluent soluble COD concentrations were significantly higher at low wastewater temperatures than higher wastewater temperatures at equal SRTs. This indicates that carbon treatment performance increases with increasing temperature (within a practical range of temperatures). A similar trend was observed in effluent COD<sub>T</sub> data for this work (Table 3). As noted, soluble COD values were not used during this study; therefore, the effluent COD<sub>T</sub> values are somewhat elevated due to the

presence of suspended solids in the effluent. A two-sample t-test was used for a statistical comparison of the mean  $\text{COD}_T$  treatment efficiencies at the different operational temperatures studied for each SRT. This type of statistical analysis was used to analyze pairs of data to determine the likelihood that the mean of one data set was statistically equal to the mean of another for a given confidence interval. Since it is assumed that the data are random and normally distributed, the distribution of the data was first analyzed according to the method described by McClave and Sincich (2000). A normal distribution of SRTs could not be rejected at a 95% confidence interval when comparing treatment efficiencies for different operating temperatures. The two-sample t-test showed that there was no statistical similarity between the  $\text{COD}_T$  treatment efficiency between the 22°C and 7°C for all SRTs. However the 22°C and the 12°C  $\text{COD}_T$  treatment efficiencies were not statistically different at a 95% confidence interval. According to these findings, carbon treatment can be accomplished at temperatures above 12°C and at an SRT of 2 days.

## **4.2 Solids Analysis Data**

The daily collection of solids data began upon reactor start-up. Figure 6 depicts reactor solids data (as MLSS) and delineates the three operational temperatures (22, 7 and 12°C) utilized during this study.

As previously discussed, initial operation of the reactor system at 22°C was interrupted by a major perturbation in wastewater strength resulting from maintenance of the anaerobic digester at the full-scale facility. The increase in wastewater strength to the bench-scale system was measured from October 20, 1999 to November 2, 1999 (day 23 to day

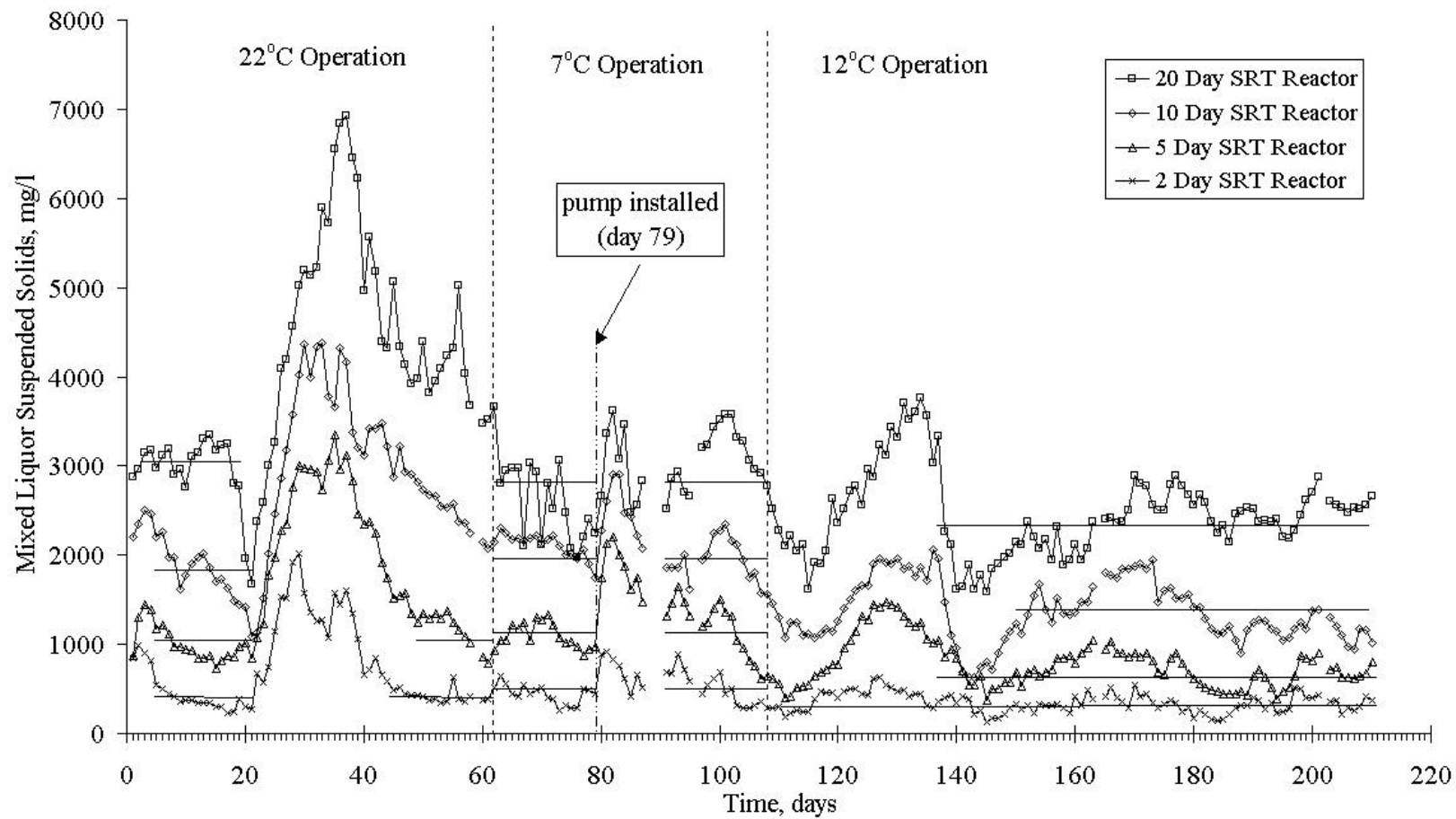


Figure 6. MLSS concentration due to changing temperature and COD concentrations throughout the treatability study.

36). A corresponding increase in MLSS was noted in each SRT reactor for a period in excess of 3 weeks. A fluctuation in MLSS also occurred during the installation of an influent circulation pump in the wastewater storage tank. The pump was installed on day 79 during 7°C operation and a subsequent increase in MLSS was observed for several days after this change in the treatment system. A fair amount of solids had collected on the bottom of the influent holding tank before the circulation pump was installed. The increase in MLSS as a result of installation of the mixing pump can be seen in Figure 6.

Excluding the two previously mentioned time periods, steady state with respect to solids (MLSS) was determined for each SRT by visual inspection of Figure 6. Stable operation is illustrated in Figure 6 by isoconcentrate lines, where the length denotes duration and the vertical position illustrates average MLSS concentration.

During operation at 22°C, the 20-day SRT system was determined to be at steady state from day 3 to day 19. The 10-day SRT system was determined to be at steady state from day 8 to day 20 and from day 56 to day 61. The 5-day SRT system was determined to be at steady state between day 5 and day 20 and day 49 and day 61. The 2-day SRT was determined to be at steady state from day 5 to day 21 and from day 44 to day 61. Based on the MLSS data during 22°C operation, the shorter SRT systems reestablished steady state much faster than did the longer SRT systems after the first perturbation.

During operation at 7°C, steady state with respect to solids was delineated as follows. The 20-day SRT was found to be at steady state from day 63 to day 78 and from day 90 to day 107. The 10-day SRT was found to be at steady state from day 63 to day 77 and from day 90 to day 107. The 5-day SRT was found to be at steady state from day

63 to day 77 and from day 97 to day 107. The 2-day SRT was found to be at steady state from day 63 to day 77 and from day 90 to day 107.

During operation at 12°C, steady state operation with respect to solids was delineated as follows. The 20-day SRT was found to be at steady state from day 137 to day 213. The 10-day SRT was found to be at steady state from day 150 to day 213. The 5-day SRT was found to be at steady state from day 138 to day 213. The 2-day SRT was found to be at steady state from day 111 to 213. Averages and standard deviations for the steady state MLSS concentrations are shown in Table 4.

The steady state averages indicate that a drop in temperature (22°C to 7°C) resulted in an increase in steady state MLSS concentration. This was unexpected since lower temperatures are commonly known to depress bacterial growth rates. Conversely, during increased operational temperature (7°C to 12°C), a decrease in MLSS was observed.

Using a two sample t-test assuming unequal variances, steady state MLSS concentration was analyzed at each temperature for all SRTs to determine whether the mean values were different. The t-test results indicated that differences in the steady state MLSS concentrations for the 20, 10 and 2-day SRTs among 22°C and 7°C operation were not statistically similar; whereas, the 5-day SRT steady state MLSS averages were statistically similar between 22°C and 7°C. In addition, average steady state influent COD<sub>T</sub> values were similar during both 22°C and 7°C operation.

Although MLSS concentration varied at different temperatures, the lowest MLSS concentrations were observed at 12°C, not at 7°C as expected.



Table 4. Average MLSS concentration during defined steady state operation.

Reactor	22°C Operation		7°C Operation		12°C Operation	
	MLSS, mg/L	Std. Dev., mg/L	MLSS, mg/L	Std. Dev., mg/L	MLSS, mg/L	Std. Dev., mg/L
20-day SRT	3081.2	185.3	2850.0	448.2	2352.6	340.2
10-day SRT	1875.0	296.8	2049.0	196.7	1362.8	269.6
5-day SRT	1042.1	194.8	1119.2	209.4	696.8	167.8
2-day SRT	392.1	89.3	477.4	156.9	345.5	108.7

Also, the steady state MLSS data from 12°C and 7°C were found to be statistically different by a two sample t-test for a 95% confidence interval of steady state data for the 20, 10 and 5-day SRTs. Since this work employed actual settled municipal wastewater, fluctuations in wastewater strength, not different operational temperatures, were thought to have contributed to the observed variations in steady state MLSS. Average influent COD<sub>T</sub> during the study were 283.2 mg/L (22°C), 272.7 mg/L (7°C) and 211.9 mg/L (12°C). The difference in average influent strength during 22°C and 7°C operation was relatively small; whereas, the average influent strength during 12°C operation was dramatically less ( $\approx$  20%) than during the other two operational temperatures. Since biomass growth is directly proportional to influent COD, lower COD levels should support fewer bacteria. However, changes in heterotrophic growth yield with temperature could also account for variations in MLSS. Randall *et al.* (1992) investigated temperature impacts on yield and determined, from bench-scale fully aerobic activated sludge systems, that the heterotrophic yield coefficient, 0.41 mg VSS/ mg COD, was fairly constant over a temperature range of 10 – 20°C. The variation in steady state MLSS concentration noted for temperature is believed due to changes in wastewater strength rather than temperature or other factors.

In order to compare MLSS data with changes in influent COD<sub>T</sub>, F/M ratios were plotted for each SRT to identify periods of relatively constant F to M.

High F/M ratios cause dispersed growth due to high substrate availability causing exponential growth; whereas, low F/M ratios are associated with low bacterial metabolism rates. Even though bacteria are considered to be in a state of endogenous growth, metabolism of organics is nearly complete and microorganisms flocculate rapidly and settle out of solution by gravity (Krishna and Loosdrecht, 1999). Hence, a constant F/M ratio would suggest a stable metabolic state and hence stable operation. Using F/M ratios as a normalizing parameter, solids steady state time periods were checked against F/M ratios to verify periods of steady state.

Figure 7 shows the F/M ratios for each SRT over the duration of the study. The F/M ratio maintained in the aeration tank defines the operation of an activated sludge system (Viessman and Hammer, 1993). During 22°C operation, the 20-day SRT had an average F/M ratio of 0.257 mg COD<sub>T</sub>/mg VSS/day with a standard deviation of  $\pm 0.1082$  (neglecting the perturbation). The 10-, 5- and 2-day SRTs had an average F/M ratio of  $0.4010 \pm 0.1467$ ,  $0.8174 \pm 0.3773$  and  $1.8660 \pm 0.8543$ , respectively. The 20- and 10-day SRTs maintained relatively stable F/M ratios with limited fluctuation during 22°C operation; whereas, the 5- and 2-day SRT F/M ratios were initially more variable. These findings did not agree with the steady state periods determined from MLSS values, which suggest that steady state for the higher SRTs was shorter than those at lower SRTs during 22°C operation. During 7°C operation, the 20-day SRT had an average F/M ratio of 0.3527 mg COD<sub>T</sub>/mg VSS/day with a standard deviation of  $\pm 0.2513$ .

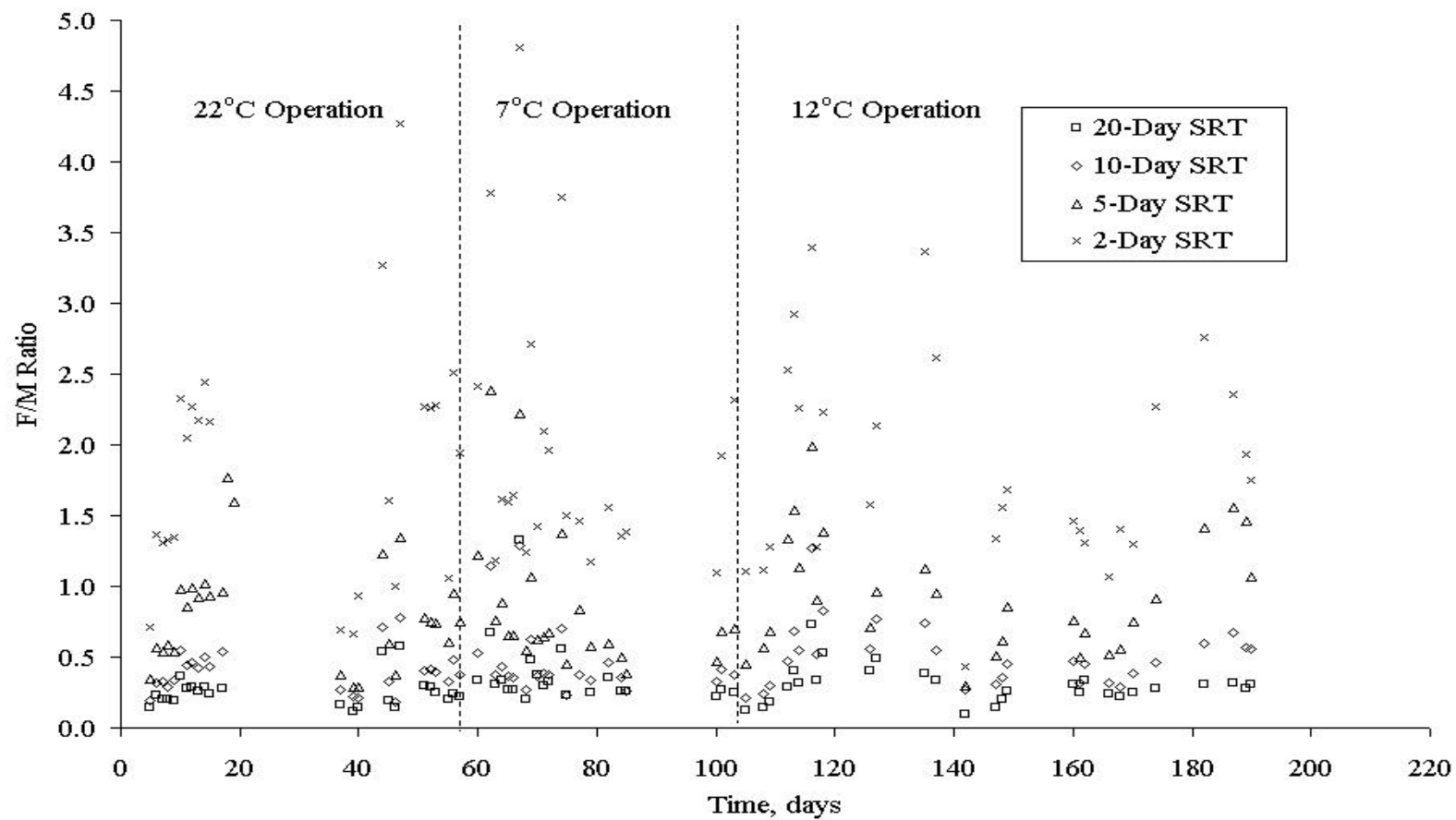


Figure 7. F/M ratios for each SRT studied during this bench-scale study.

The 10-, 5- and 2-day SRT had an average F/M ratio of  $0.4463 \pm 0.2683$ ,  $0.8132 \pm 0.5182$  and  $1.9046 \pm 0.9800$ , respectively. F/M ratios for all SRTs during this operational period were more variable than those measured at 22 and 12°C based on calculated standard deviations. This, in conjunction with poor  $\text{COD}_T$  removal efficiencies, suggests that steady state with respect to solids was not achieved for any SRT studied during 7°C operation.

During 12°C operation, F/M ratios for all SRTs were least variable among the three temperature regimes studied but were slightly higher than those observed at 22°C operation. During this operational period, F/M ratios for the 20-, 10-, 5- and 2-day SRTs were  $0.3158 \pm 0.1270$ ,  $0.5273 \pm 0.2181$ ,  $0.9674 \pm 0.4134$  and  $1.9068 \pm 0.7304$ , respectively. The 20- and 10-day SRT F/M ratios were relatively stable during operation at 12°C, whereas the 5- and 2-day SRT F/M ratios were more variable. Again, these findings contradict the previously determined solids steady state periods that suggest the 2-day SRT system was at steady state very soon after the temperature shift to 12°C. This comparison shows that without stable influent substrate, solids steady state for this bench scale system cannot be defined based solely on stable solids concentration within the reactor. Caution is also warranted when using the F/M ratios presented since a gross measure of influent carbon substrate was used in the analysis rather than the BOD5 recommended by Krishna and Looschrecht (1999).

The MLVSS concentrations were assessed throughout the treatment study. The volatile fraction of the MLSS (MLVSS) represents the fraction of solids which oxidizes to carbon dioxide ( $\text{CO}_2$ ) when ignited at 550°C. The measured MLVSS values for each SRT proved to be a consistent fraction of the MLSS for the entire duration of the

experiment. For the 20-day SRT, the percentage of MLVSS to MLSS was 73% with a standard deviation of 3%. For the 10-day SRT, the percentage of MLVSS to MLSS was 76% with a standard deviation of 4%. For the 5-day SRT, the percentage of MLVSS to MLSS was 79% of the MLSS with a standard deviation of 6%. Finally for the 2-day SRT, the percentage of MLVSS to MLSS was 83% with a standard deviation of 9%. A decline in % of total solids as volatile solids was observed with a decline in operational temperature during this study. Data is summarized in Table 5. A two sample t-test assuming unequal variances was performed to determine whether two sample means are statistically similar. In this case, steady state MLVSS concentrations were compared between SRT at each temperature studied. The 20-day and 10-day SRT MLVSS values were found to be not statistically different for a 95% confidence interval during 22°C operation. At 12°C, MLVSS values between the 20- and 10-day, 10- and 5-day, and the 5- and 2-day SRT systems were found to be statistically similar. No statistical similarity was determined using the two-sample t-test assuming unequal variances for steady state MLVSS data collected during 7°C operation; even though the averages presented in Table 7 to seem to overlap when considering their standard deviations. This analysis shows that at 22°C higher SRTs had similar MLVSS while low SRT MLVSS values were similar but distinctly different from the higher SRTs studied. For 7°C operation this relationship was not observed showing a more distinct separation of MLVSS for the SRTs studied.

### **4.3 Biosolids Settling**

Sludge volume index, SVI, indicates the settlability of biosolids. SVI data are evaluated using previously suggested guidelines, where excellent, moderate and poor

Table 5. Average percent MLVSS during defined steady state operation.

Reactor	22°C Operation		7°C Operation		12°C Operation	
	MLVSS, %	Std. Dev., %	MLVSS, %	Std. Dev., %	MLVSS, %	Std. Dev., %
20-day SRT	76.4	1.37	73.7	2.36	74.3	1.68
10-day SRT	77.3	2.26	74.9	1.54	78.8	2.99
5-day SRT	81.2	3.41	77.6	3.31	82.0	5.58
2-day SRT	88.9	4.50	81.3	5.54	84.5	5.87

settling are represented by SVI values < 80 mL/g, 80-150 mL/g and > 150 mL/g, respectively (Grady *et al.*, 1999). SVI data for this study are presented in Figure 8.

During start-up operation (22°C), SVI values for each SRT were excellent (below 80 mL/g). However, after one week, SVI values increased dramatically for the 10-, 5- and 2- day SRTs, however, SVI values for the 20-day SRT remained in the moderate range. During the initial period of operation, before the perturbation event, the 5- and 2- day SRT system SVIs followed similar trends of increase reaching maximum values of 1212 mL/g (day 15) and 1468 mL/g (day 17), respectively. The 10-day SRT system also experienced a similar increase in SVI (maximum 715 mL/g on day 21). The 20-day reactor reached a maximum SVI of 135 on day 17 during initial operation, which proved to be good compared to the other maximum SVI values observed during initial operation. Settling improved in all reactors during the period of high strength wastewater feed (day 23 to day 36). By day 40 of operation, SVI values for the 10-, 5- and 2-day SRT achieved minimum values of 43, 150 and 133 mL/g, respectively. These minimum SVI values occurred approximately 17 days after the introduction of high strength wastewater.

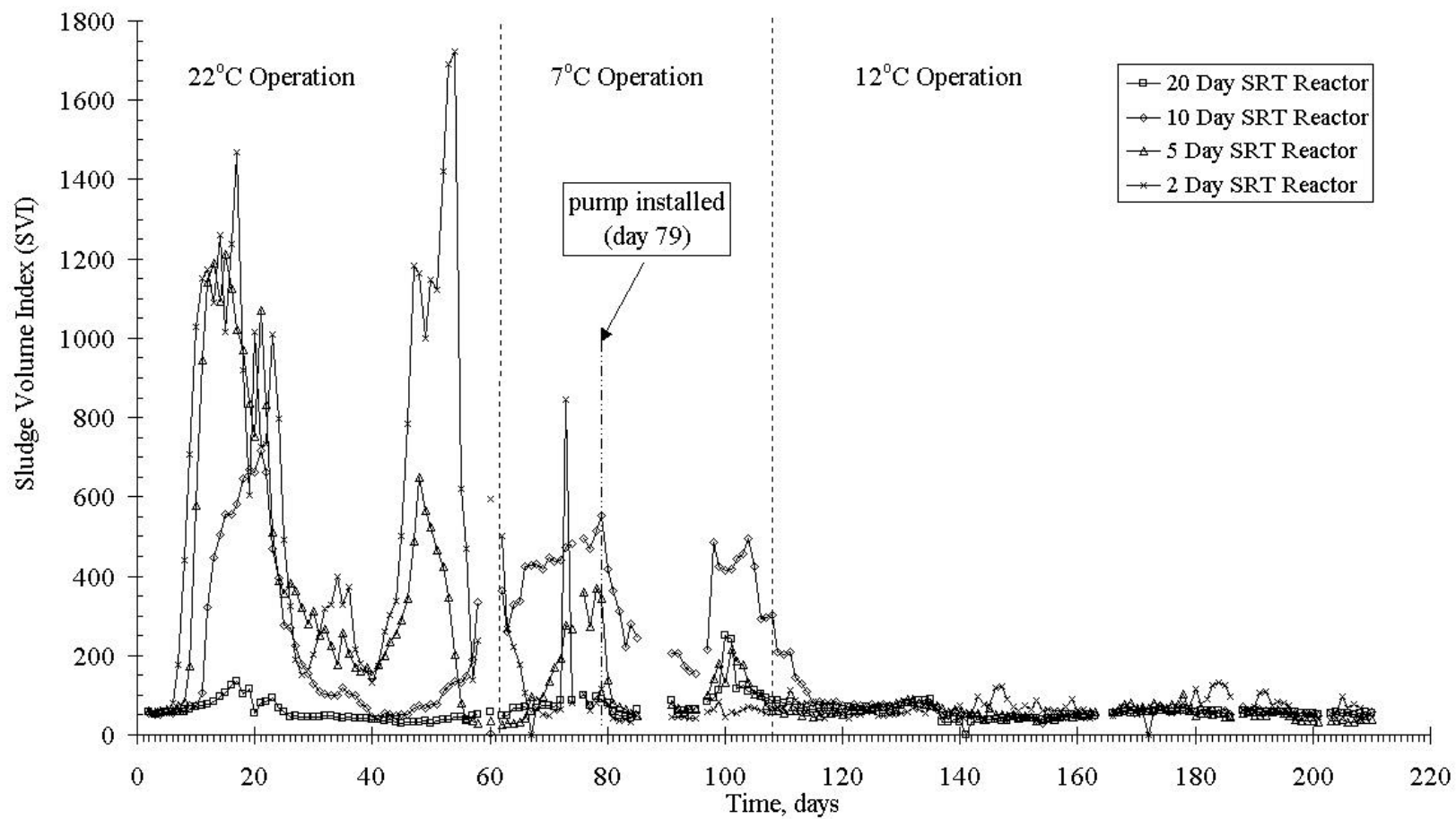


Figure 8. Effect of temperature (22°C to 7°C) on settling performance throughout the treatability study.

However, improved settling characteristics were not maintained once the wastewater feed returned to normal strength. There may be a relationship between increasing SRT and improved settling of mixed-liquor during reactor start-up (i.e. biosolids acclimation). The increase in wastewater strength was a major factor, which impacted recorded SVI values. This impact was observed to increase with decreasing SRT. This increase in wastewater strength can be translated into an increase in organic loading or F/M ratio. The F/M ratios for each SRT were determined to identify if this may have influenced biosolids settling.

The F/M ratio maintained in the aeration tank can be used to define the operation of an activated sludge system (Viessman and Hammer, 1993; Krishna and Loosdrecht, 1999). As discussed previously, low SRTs, where microorganisms are in the exponential growth phase, the F/M ratio is high. This creates an environment which favors dispersed growth and poor biomass settling. Higher organic loading can also lead to higher ESS concentrations through the bulking phenomena (Churchwell *et al*, 1980). However, when the F/M ratio is low (high SRTs), metabolic activity is considered to be in the endogenous phase of growth. Although the rate of metabolism is low, degradation of organic substrate is nearly complete during this stage and microorganisms flocculate rapidly and settle out of solution by gravity. Thus, SRT can be an indicator for proper biosolids settling. During 22°C operation, the F/M ratio (based on solids steady state periods) increased for the 20 and 10-day SRTs from 0.247 and 0.438 mg COD<sub>T</sub>/mg MLVSS/day to values of 0.470 and 0.690 mg COD<sub>T</sub>/mg MLVSS/day during the high strength wastewater perturbation. While the 5 and 2-day SRTs had lower F/M ratios (0.895 and 1.689 mg COD<sub>T</sub>/mg MLVSS · day) during the perturbation as compared to



steady state operation (0.901 and 2.02 mg COD<sub>T</sub>/mg MLVSS · day) at 22°C. This suggests that the low SRTs were already experiencing high organic loading which contributed to poor biosolids settling. The F/M values for steady state and the perturbation for the 5-day SRT are relatively close, which does not explain the improvement in settling between these two periods. Churchwell et al (1980) prevented sludge bulking and associated settling problems by controlling organic loading at an approximate level of 0.2 kg COD applied per day per 0.5 kg MLVSS (0.4 mg COD<sub>T</sub>/mg MLVSS · day).

Since settling efficiency can vary with SRT, a two-sample t-test analysis was conducted to assess the impact of SRT on biosolids settling. With the 20-day SRT achieving the best settling characteristics out of all SRTs studied, the 20-day SRT was compared to the 10, 5 and 2-day SRTs at 12°C. 12°C was chosen due to the efficient settling observed at all SRTs and therefore eliminating possible disturbances, which may provide inaccurate results. The average SVI values for the 20 and 5-day SRT reactors for the 12°C steady state time periods were found to be statistically similar for a confidence interval of 95%. No statistical similarity was found between the 20-day SRT steady state SVI values and the 10- and 2-day data during 12°C. However, the SVI values between the 10- and 5-day SRT systems were found not statistically different for a confidence interval of 95% by the two-sample t-test assuming unequal variances. These findings indicate that longer SRTs (low F/M ratios) promote more efficient settling. Similar results were found by Parker (2001) when conducting a bench-scale experiment on DO and SRT effects on activated sludge performance.

The change in temperature from 22°C to 7°C was accompanied by a dramatic decrease in the SVI values for the 2-day SRT (Table 6). The SVI values before the temperature decrease were poor for the 2- and 10-day SRT reactors. Whereas after the temperature decrease, the 10-day SRT reactor SVI values remained poor but the 2-day SRT reactor SVI values improved. The 5-day SRT reactor SVI values increased to poor levels within a week after the temperature change. During reactor operation at 7°C, a mixing pump was installed in the influent reservoir in order to homogenize the wastewater, denoted by a dotted line (day 79). From inspection of Figure 7, both the 10 and 5-day SRTs improved dramatically after installation of the influent mixing pump. For the remainder of 7°C operation, one more fluctuation in SVI was observed in the 20, 10 and 5 day SRTs, with the 10-day SRT being the most pronounced. SVIs for all SRTs were excellent after the change in temperature from 7°C to 12°C. Good settling continued for the duration of the study.

SVI values for each SRT and F/M ratios are plotted over time to illustrate the effect of F/M ratio on sludge settlability (Figures 9 through 12). The SVI and F/M ratios measured for the 20-day SRT were constantly low during the entire course of this study.

Table 6. Average SVI values during defined steady state operation.

Reactor	22°C Operation		7°C Operation		12°C Operation	
	SVI, ml/g	Std. Dev., ml/g	SVI, ml/g	Std. Dev., ml/g	SVI, ml/g	Std. Dev., ml/g
20-day $\theta_c$	81.6	26.6	94.0	45.8	51.1	9.3
10-day $\theta_c$	349.0	243.0	371.0	111.6	54.8	8.8
5-day $\theta_c$	521.0	438.7	144.9	86.1	52.0	14.1
2-day $\theta_c$	857.0	457.6	102.0	150.7	68.8	21.0

Also, the F/M ratios measured for the 10-, 5- and 2-day SRT were expectedly higher than the 20-day SRT since the magnitude of the F/M ratio is SRT dependent as can be observed in Figures 9 through 12. The SVIs for the 10-, 5- and 2-day SRTs were poor during start-up (22°C operation) and temperature considerations during 7°C may have contributed, in part, to the continuation of poor settling at these SRTs. However, SVIs in the 10-, 5- and 2-day SRTs achieved levels denoting excellent settling. This was in spite of drastically different F/M ratios measured during the latter part of operation. The excellent settling in combination with low F/M ratios observed in the 20-day SRT supported settling theory identified in literature and also reinforced observed trends in SVI as improving with increasing SRT (lower F/M ratio). However, SVIs at all SRT during the final stage of operation were excellent, denoting that reactor acclimation, apart from F/M ratios, was also an important factor in achieving a proper settling sludge.

Since the literature indicated that settling performance is a function of temperature, a two-sample t-test was performed on all SRTs for each temperature studied during this research. The 20-day SRT average SVI values for 22°C and 7°C operation were not statistically different at a confidence interval of 95% where average SVI values for 22°C and 12°C were statistically different at a 95% confidence interval. Also, using the two-sample t-test, the 20-day SRT system steady state SVI values collected at 7°C and 12°C were not statistically different for a 95% confidence interval. This correspondence was also identified for the 10-, 5- and 2-day SRT reactors. Also, independently the 10- and 5-day SRT reactor steady state SVI values were not statistically different for a 95% confidence interval for data collected at 22°C and 12°C. The 10-day SRT system average steady state SVI values were not statistically different at

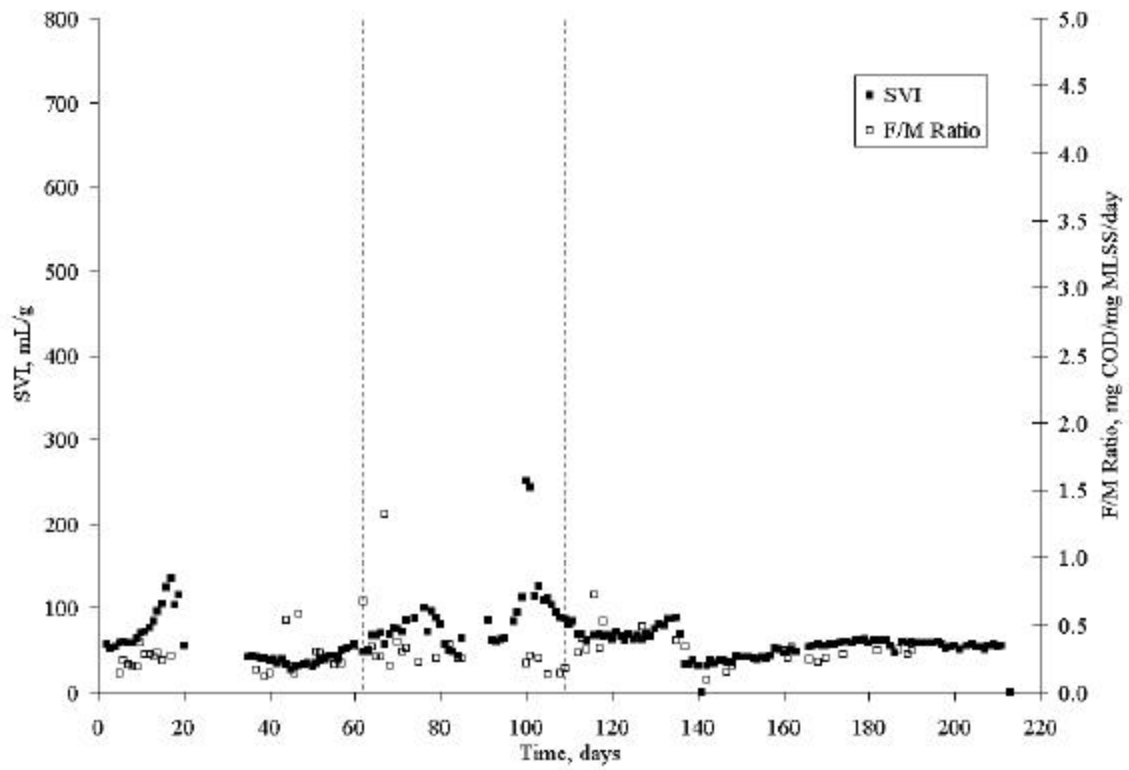


Figure 9. A comparison of SVI and F/M ratios for the 20-day SRT during this study.

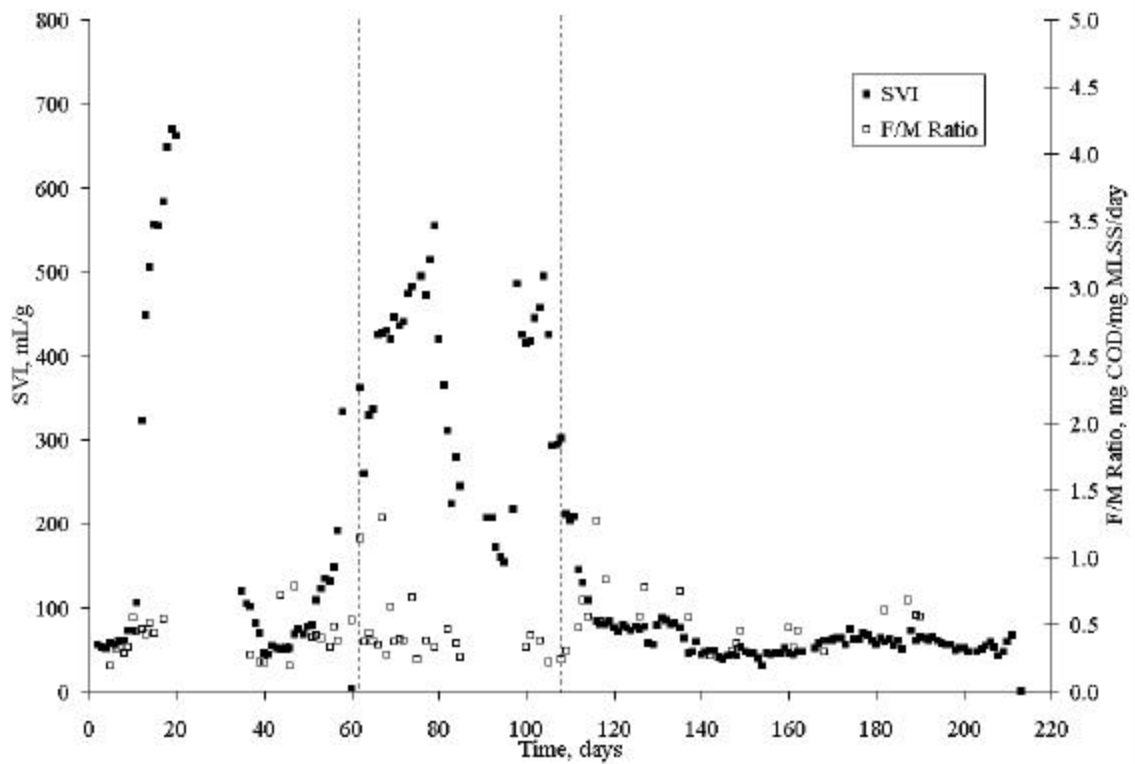


Figure 10. A comparison of SVI and F/M ratios for the 10-day SRT during this study.

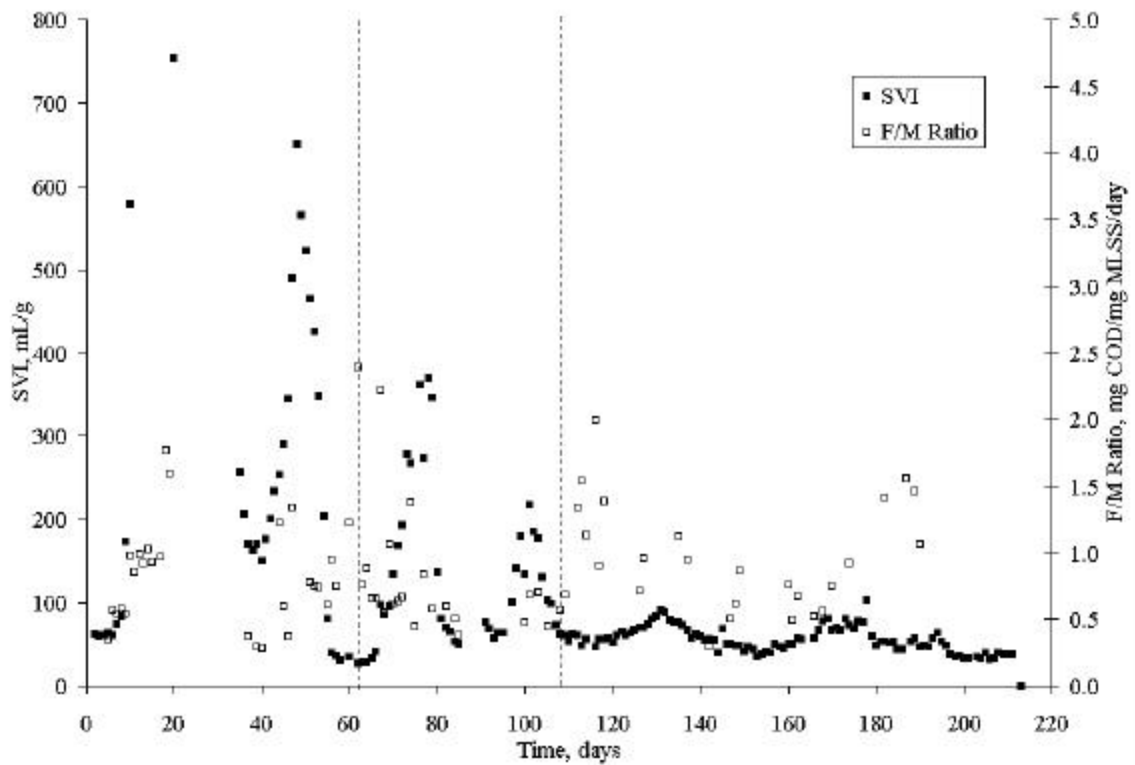


Figure 11. A comparison of SVI and F/M ratios for the 5-day SRT during this study.

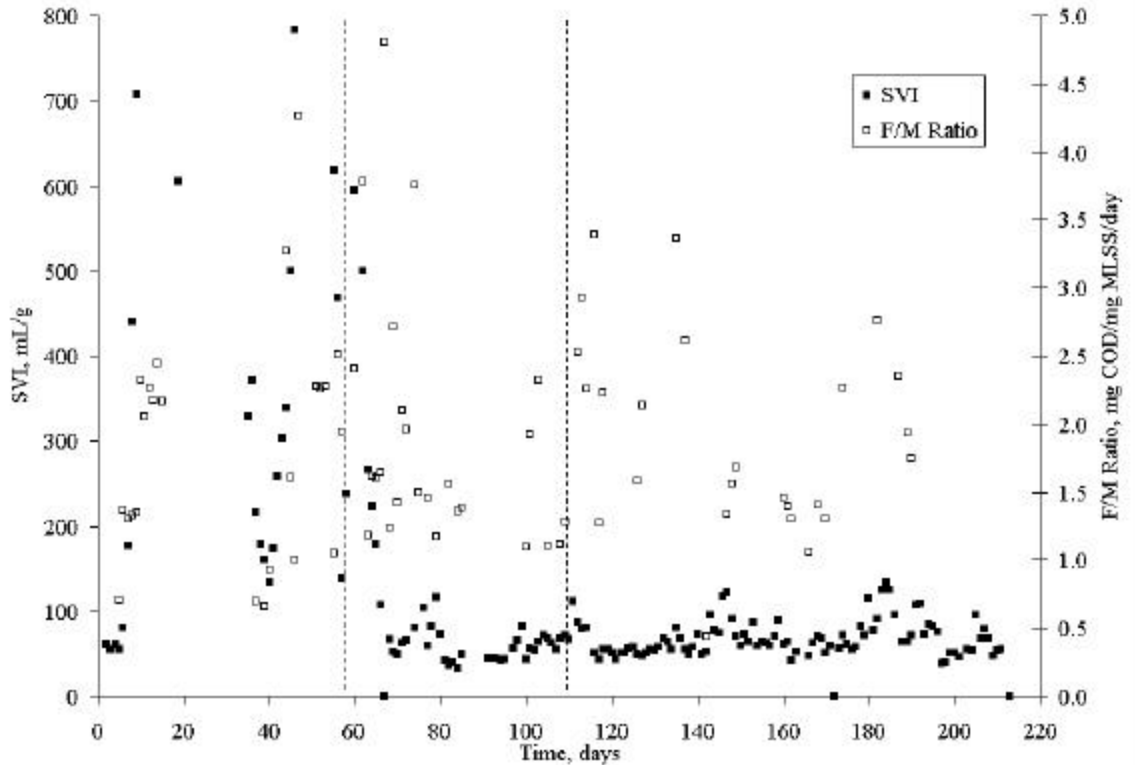


Figure 12. A comparison of SVI and F/M ratios for the 2-day SRT during this study.

a confidence interval of 95% between 22 and 7°C operation. The 5-day SRT reactor did not show statistical similarity for average SVI values when temperature regimes were compared. However, the 2-day SRT average steady state SVI values for 7°C and 12°C were not to be statistically different at a confidence interval of 95%, where a comparison of SVIs between 22 and 7°C and between 22 and 12°C were determined to be statistically different at a confidence interval of 95%.

Krishna and Loosdrecht (1999) determined that temperature effected SVI in a sequencing batch reactor treatment system. At a temperature of 15°C, an SVI of 110 ml/g was measured whereas at a temperature of 25°C, an SVI of 200 ml/g was noted. The researchers concluded that solids settlability, expressed as SVI, strongly decreased with increasing temperature, and that biosolids settleability in systems with a low SRT was significantly influenced by temperature, with an increased SVI at higher temperature. The determined statistical similarity of average SVI values did not follow an identifiable trend with respect to SRT or temperature. However, the consistent improvement of SVI during the course of this study indicate that biomass acclimation, not temperature or SRT, is the determining factor for low SVIs.

#### **4.4 Effluent Suspended Solids**

In the activated sludge process, it is necessary to separate the treated wastewater from the biomass, thereby producing a clear effluent (Fuchs and Staudinger, 1999). The effluent suspended solids (ESS) values reflect external clarifier performance as well as settling characteristics of the biosolids. The ESS data show that overall external clarifier performance of the final design model was satisfactory (Figure 13). After initial testing

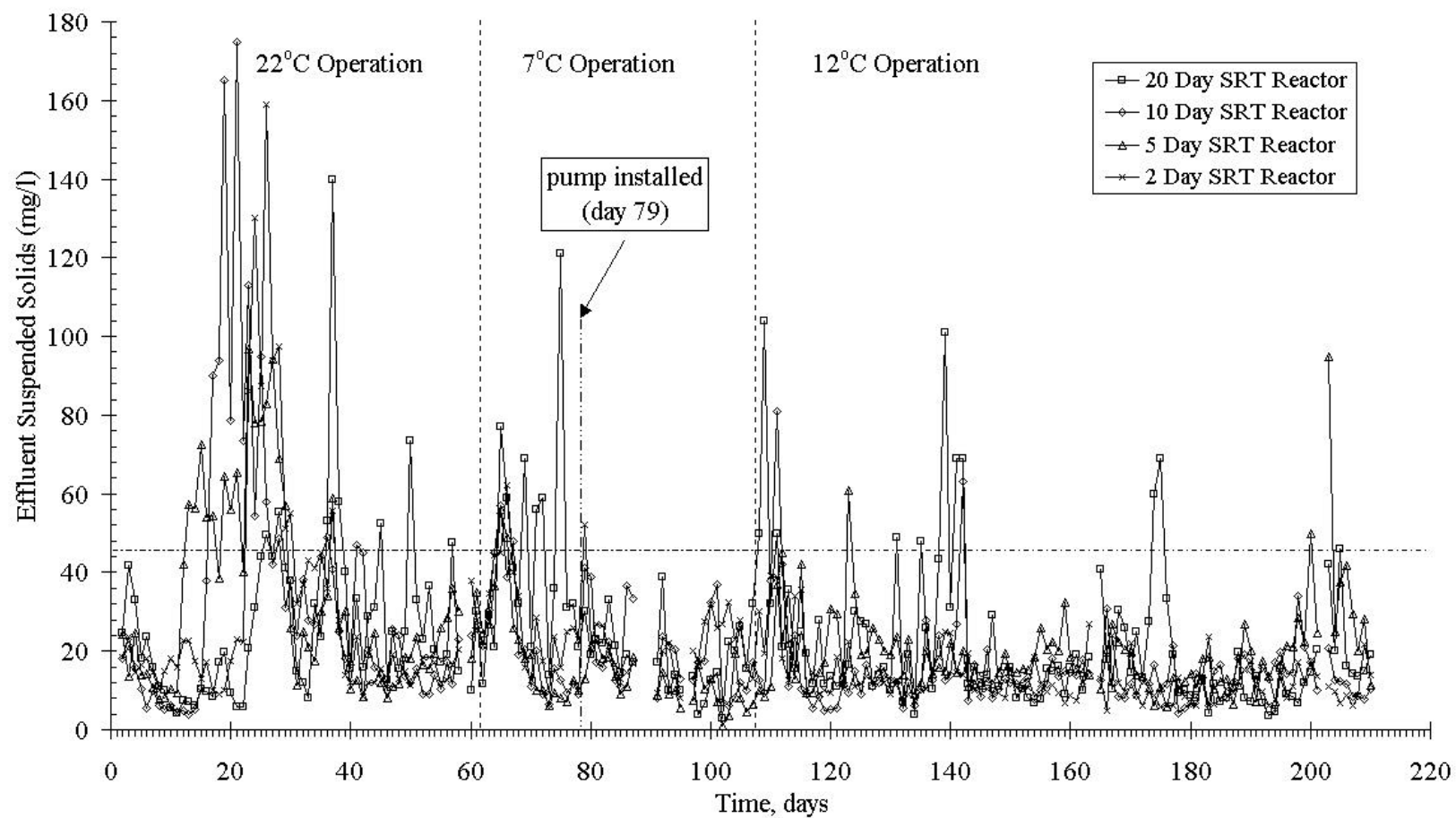


Figure 13. Effluent suspended solids (ESS) concentration throughout the treatability study.

of the custom external clarifier, several design changes were made to improve performance. The major changes included an increased diameter underflow collection area to prevent bridging of collected solids and improve sludge thickening. Also, the addition of a mechanical scraper arm proved useful in preventing attached growth on the inner vertical walls and in facilitating the movement of settleable solids to the biosolids underflow port. The design changes were made before start-up and operation of treatment systems used in this study. Based on the ESS data, the changes did prove useful and successful. The increased underflow port provided more room for solids compaction which seemed to aid in return sludge flow. The scraper arm did prevent large amounts of solids from adhering to the clarifier wall; however, minor adjustments in the shape and angle of the scraper arm were made to ensure proper contact between the clarifier wall and the scraper arm. Quiescent conditions within the clarifier were maintained during scraper arm operation; however, periodical manual cleaning was necessary to remove solids attached to the scraper arm assembly itself. Proper clarifier performance was integral in stable operation of the bench-scale treatment system.

During this study, ESS data display several high discharge events that correlated with changes in operational conditions (Figure 13). These events were characterized by an increase in ESS for all reactor systems. However, for the majority of operation the ESS was below 45 mg/L, the maximum daily discharge limit for KUB's Kuwahee WWTP. During initial operation at 22°C, ESS values for all SRTs were excellent; however, concentrations increased during the high strength wastewater event for the entire system. During this perturbation, the peak ESS values for the 20-, 10-, 5- and 2-day systems were 55.4, 113.1, 96.7 and 158.9 mg/L, respectively. These high ESS



concentrations were attributed to the explosion of biological growth that accompanied the high-strength wastewater. ESS values dropped well below the 45 mg/L target after the perturbation (day 40) with occasional exceptions during 22°C operation.

The temperature shift from 22°C to 7°C was accompanied by an increase in ESS within 3 to 4 days after the temperature shift for all SRTs. The 20-, 10-, 5- and 2-day SRT systems reached ESS concentrations of 77, 57, 56 and 62 mg/L, respectively, after the temperature change. Whereas, the ESS values the day of the temperature shift were 11.5, 21.5, 24.5 and 21.2 mg/L for the 20-, 10-, 5- and 2-day systems, respectively. ESS concentrations returned to values below 30 mg/L within a day or two after the event and remained within EPA guidelines (< 30 mg/L on average) throughout the remainder of 7°C operation. However, there was a second increase in ESS during 7°C operation which corresponded with the installation of the influent feed recirculation pump on day 79. Although ESS levels fluctuated during 7°C operation, steady state values remained near or below the 30 mg/L effluent discharge level for each SRT (Table 7). Effluent SS levels were also impacted by the second temperature shift (7°C to 12°C) on January 13, 2000 (day 108). ESS concentrations increased to maximum levels within approximately 3 to 4 days of the temperature change but quickly dropped to low levels. This temporal trend was similar for each ESS event identified.

Table 7. Average ESS concentrations during defined steady state operation.

Reactor	22°C Operation		7°C Operation		12°C Operation	
	ESS, mg/L	Std. Dev., mg/L	ESS, mg/L	Std. Dev., mg/L	ESS, mg/L	Std. Dev., mg/L
20-day $\theta_c$	14.6	10.5	30.6	25.1	20.1	17.9
10-day $\theta_c$	34.1	44.3	19.9	13.3	13.5	5.3
5-day $\theta_c$	30.5	19.4	16.0	13.6	18.7	12.1
2-day $\theta_c$	16.6	5.7	24.8	12.0	12.6	6.4

## 4.5 Nitrification Performance Data

### 4.5.1 Ammonia removal performance

Ammonia removal was quantified as the difference between influent and effluent ammonia (mg/L  $\text{NH}_4^+-\text{N}$ ) concentrations. Complete ammonia removal was achieved when the effluent ammonia concentration was less than 0.1 mg/L, a restriction that represented the detection limit of the ammonium ion selective electrode. Operational influent and effluent ammonia data are presented here in Figure 14.

The average influent ammonia concentration, neglecting the perturbation, was  $23.3 \pm 1.3$  mg/L as N ( $\pm$  represents standard deviation at 95% confidence interval about the mean for the following discussion) at 22°C. Complete ammonia removal was observed at 20-, 10- and 5-day SRT systems directly after initial startup. The 2-day SRT reactor, however, did not achieve complete ammonia removal until after the perturbation (day 39). During the perturbation, the average ammonia concentration was  $41.6 \pm 3.6$  mg/L as N. This constitutes approximately a two-fold increase in the influent ammonia concentration.

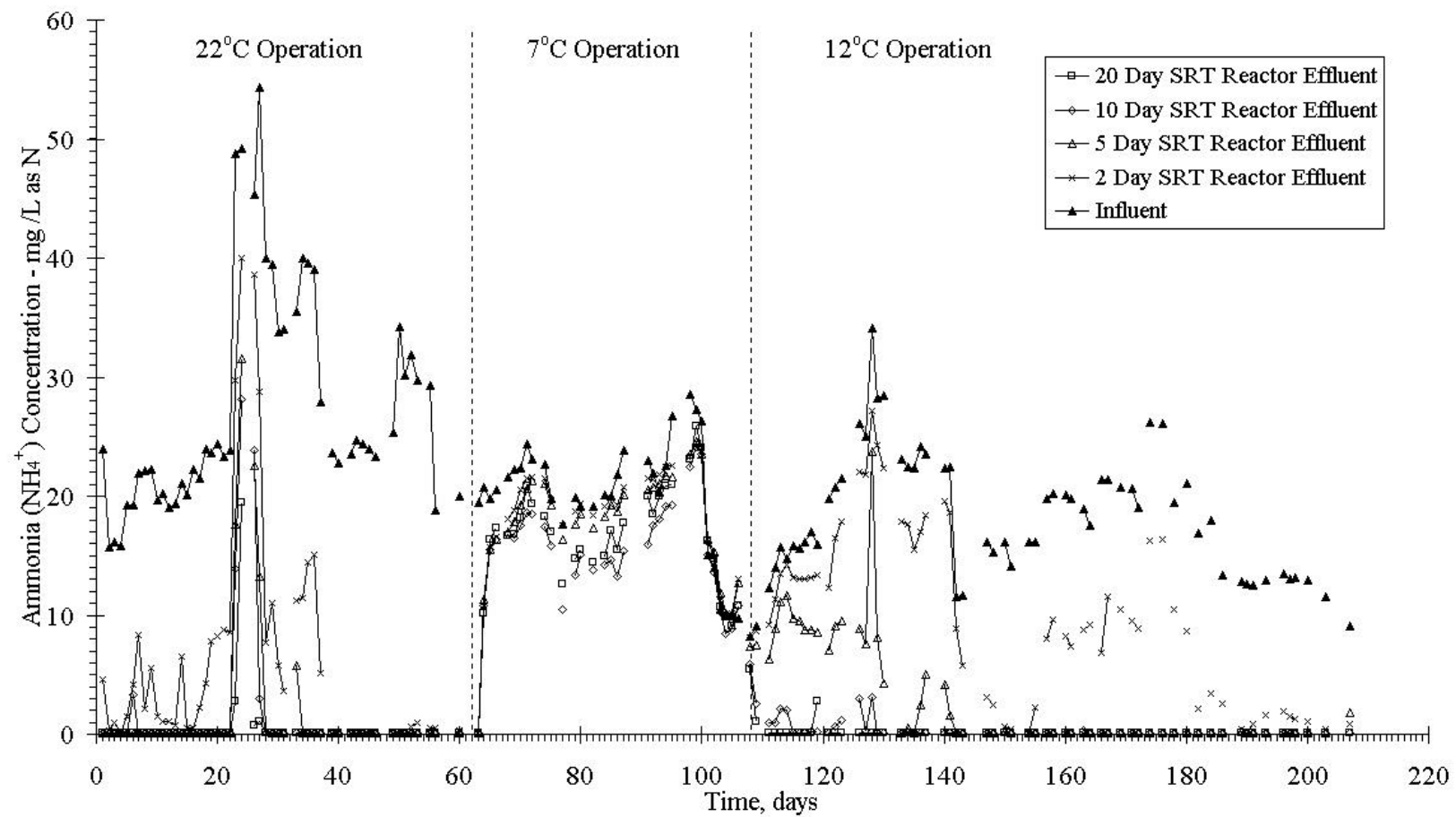


Figure 14. Ammonia removal as a function of temperature over the course of the treatability study.

During this time period, the effluent ammonia concentrations were  $2.1 \pm 3.1$ ,  $5.8 \pm 5.8$ ,  $7.7 \pm 6.2$  and  $18.1 \pm 7.2$  mg/L as N for the 20-, 10-, 5- and 2-day SRT systems. This increase in wastewater strength was accompanied by a loss in ammonia treatment efficiency, which varied according to SRT. The corresponding ammonia removal efficiencies during the perturbation were  $95.7 \pm 6.4$ ,  $87.8 \pm 12.2$ ,  $83.9 \pm 12.9$ , and  $59.2 \pm 13.8$  percent, for the 20-, 10-, 5- and 2-day SRTs, respectively.

Stable operation for the 20-day SRT system was reestablished within four days after the event had occurred. Both the 10- and 5-day SRTs regained efficient ammonia removal within five days after the perturbation, whereas the 2-day SRT reactor required an additional 10 days before stable operation was reestablished. All SRTs achieved stable operation (complete ammonia removal ( $< 0.1$  mg/L as N)) at  $22^{\circ}\text{C}$  from day 39 to day 60, constituting 22 days of stable ammonia removal at  $22^{\circ}\text{C}$ . The influent ammonia concentration during this time frame was  $27.7 \pm 2$  mg/L as N.

The first temperature shift (to  $7^{\circ}\text{C}$ ) occurred on day 62. Within twelve hours after the temperature adjustment, the reactor contents were at  $7^{\circ}\text{C}$ . The decrease in temperature was accompanied by a loss in ammonia removal efficiency. A reduction in ammonia treatment occurred rapidly and within 3 days of the temperature shift, near influent ammonia concentrations were measured in the effluent at each SRT. The average influent ammonia concentration during  $7^{\circ}\text{C}$  operation was  $19.8 \pm 1.8$  mg/L-N. Corresponding average effluent ammonia levels were 16.0 mg/L as N (20-day), 14.9 mg/L as N (10-day), 17.3 mg/L as N (5-day) and 17.7 mg/L as N (2-day). Resulting

average ammonia removal efficiencies for the 20-, 10-, 5- and 2-day SRTs at 7°C were 18, 23, 11 and 9 percent, respectively.

Although nitrate data was not available during the entire study, effluent nitrate data available during 7°C operation (day 86 to day 102) revealed that some nitrification was occurring. Average nitrate levels were 7.83 mg/L (20-day), 7.25 mg/L (10-day), 6.28 mg/L (5-day) and 7.25 mg/L (2-day) on day 86. By day 102, nitrate levels decreased to below detection levels. The decline in nitrate was accompanied by a similar decline in influent ammonia concentration. During 7°C operation, dilute wastewater was collected from the Kuwahee WWTP on day 101 for use as the influent feed for the bench-scale system. This low strength wastewater seemed to exacerbate the poor removal efficiencies already observed. Negative ammonia removal efficiencies were recorded during the dilute wastewater feed period at all SRTs. This phenomenon was ascribed to the process of ammonification which occurs when organically bound nitrogen is converted to ammonia during heterotrophic carbon utilization. The accumulation of ammonia in the effluent in excess of the influent ammonia concentration was reflected by negative ammonia removal efficiencies.

The temperature shift from 7°C to 12°C occurred on day 107. After this increase in temperature, ammonia removal improved for all SRTs; however, the speed of recovery varied according to SRT. The average influent ammonia concentration during 12°C operation was  $18.2 \pm 1.3$  mg/L-N. Complete ammonia removal was achieved in the 20-, 10-, and 5-day SRT reactors on day 121, day 129 and day 142, respectively. The 2-day SRT did not achieve consistent effluent ammonia concentrations below 0.1 mg/L-N before system shutdown (day 213); however, from day 182 until system shutdown the 2-

day SRT averaged  $90 \pm 3$  % ammonia removal efficiency. This time frame constituted the most efficient removal for the 2-day SRT during 12°C operation. Complete ammonia oxidation was not anticipated in the 2-day reactor, since this SRT approaches the minimum value required for complete ammonia oxidation.

A number of studies have indicated that temperature can have a major impact on ammonia removal during activated sludge treatment. There is a pronounced decrease in nitrification at temperatures below 18°C with little autotrophic growth occurring below 5°C (Ford, 1980). McClintock et al. (1992) showed partial ammonia removal at an SRT of 1.5 days at 20°C and 5 days at 10°C. High degrees of nitrogen removal (60-70%) were achieved at 20°C for all SRTs of 2.7 days or more. Greater removals were achieved at a 15-day SRT than at a 5-day SRT for all temperatures (McClintock et al., 1992). Since it has been documented that little or no nitrification occurs at or below 5°C, it was anticipated that effective ammonia removal would not occur at the low temperature (7°C) portion of this study. However, Tendaj *et al.* (1992) reported that when temperature remains constant at a low level over a long period of time, nitrification gradually increases. Hoilijoki *et al.* (2000) studied the effect of low temperatures on nitrification for anaerobically pretreated landfill leachate using the activated sludge process and found that at a loading rate of 0.027 g  $\text{NH}_4^+\text{-N/MLVSS/day}$  and an HRT of 3 days, complete nitrification was possible at 10°C. The researchers found that at 7°C and a loading rate of 0.023 g  $\text{NH}_4^+\text{-N/MLVSS/day}$ , ammonia removal was only 93%, and that at 5°C and a loading rate of 0.011 g  $\text{NH}_4^+\text{-N/MLVSS/day}$ , only 61% ammonia removal was observed. Oleszkiewicz (1988) studied the effect of low temperatures on nitrification/denitrification using sequencing batch reactors (SBRs) and found that the response to a decrease in

temperature was discontinuous. A more pronounced effect of temperature on nitrification was reported for the temperature range between 2–7°C. The author did document the feasibility of nitrification down to 2°C for SBRs although steady 90% nitrification efficiency at 2°C could only be accomplished at an SRT above 30 days accompanied by a significant decrease in the F/M load.

#### **4.5.2 Examination of steady state for nitrification**

Steady state operation with respect to nitrification was indirectly determined through a measured reduction in influent ammonia concentration. As discussed in the previous section, nitrogen data were not complete during the entire duration of this study. However, certain operational time frames were identified where complete ammonia removal was determined. Figure 15 presents the nitrogen speciation plot for the 20-day SRT over the entire duration of this study.

During 22°C operation, influent  $\text{NH}_4^+$ -N and effluent  $\text{NO}_3^-$ -N concentrations were quite similar. The average nitrate concentration for the 20-day SRT system was  $26.4 \pm 4.5$  mg/L as N. For the time period during which nitrate data was available (day 1 to day 28), the corresponding average ammonia concentration was  $25.8 \pm 4.2$  mg/L as N. Average nitrate concentrations were located within the 95% confidence interval for average ammonia concentration suggesting that complete nitrification did occur. On several occasions, the average nitrate level was greater than the average ammonia concentration. As discussed previously, this phenomenon was attributed to the conversion of organic nitrogen (which is not measured by the ion selective probe) to nitrate after nitrification. Neglecting the perturbation in wastewater strength, steady state

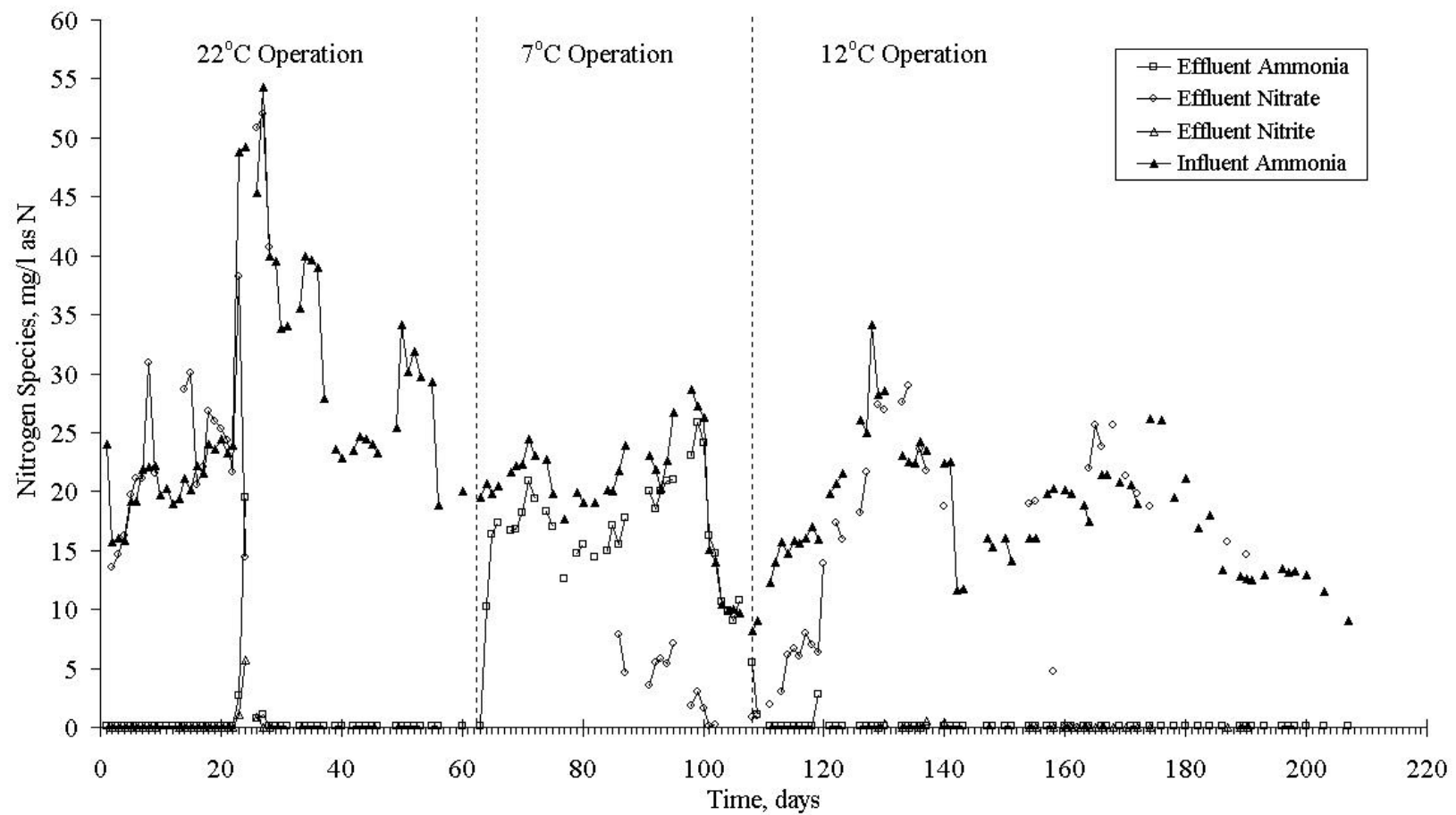


Figure 15. Nitrogen speciation plot for the 20-day SRT indicating the occurrence of complete nitrification.



operation with respect to nitrification for the 20-day SRT reactor was marked from day 1 to day 22 and from day 39 to day 60.

During 7°C operation, ammonia removal was consistently poor over the entire length of operation. Data collected during the latter part of low temperature operation showed that a moderate amount of nitrate was present (day 86 to day 95) indicating partial nitrification. Nitrate levels subsequently decreased and negative removal efficiencies were recorded during the period of low strength wastewater feed (day 104 to day 109). The build up of ammonia in the effluent to concentrations greater than those measured in the influent was attributed to ammonification. No steady state was reached during operation at 7°C.

Complete ammonia removal was reestablished at 12°C within 3 days after the temperature was increased (day 111). With the exception of day 119, effluent ammonia levels were below the detection limit (0.1 mg/L-N) during 12°C operation. Nitrate data provided additional evidence that complete nitrification was reestablished by day 129. Effluent nitrate-N and influent ammonia-N were in close proximity after this date indicating steady state operation from day 129 to day 207.

Figure 16 presents the nitrogen speciation plot for the 10-day SRT system. The average nitrate concentration during 22°C operation was  $23.0 \pm 3.7$  mg/L as N whereas the average ammonia concentration was  $27.4 \pm 2.6$  mg/L as N. Initial operation showed that complete nitrification had occurred based on low effluent ammonia (0.1 mg/L  $\text{NH}_4^+$ -N) and effluent nitrate concentrations which were similar to influent ammonia nitrogen. Nitrite was detected in small amounts during the perturbation event; however, effluent nitrate-N and influent ammonia-N levels were also in similitude after the perturbation

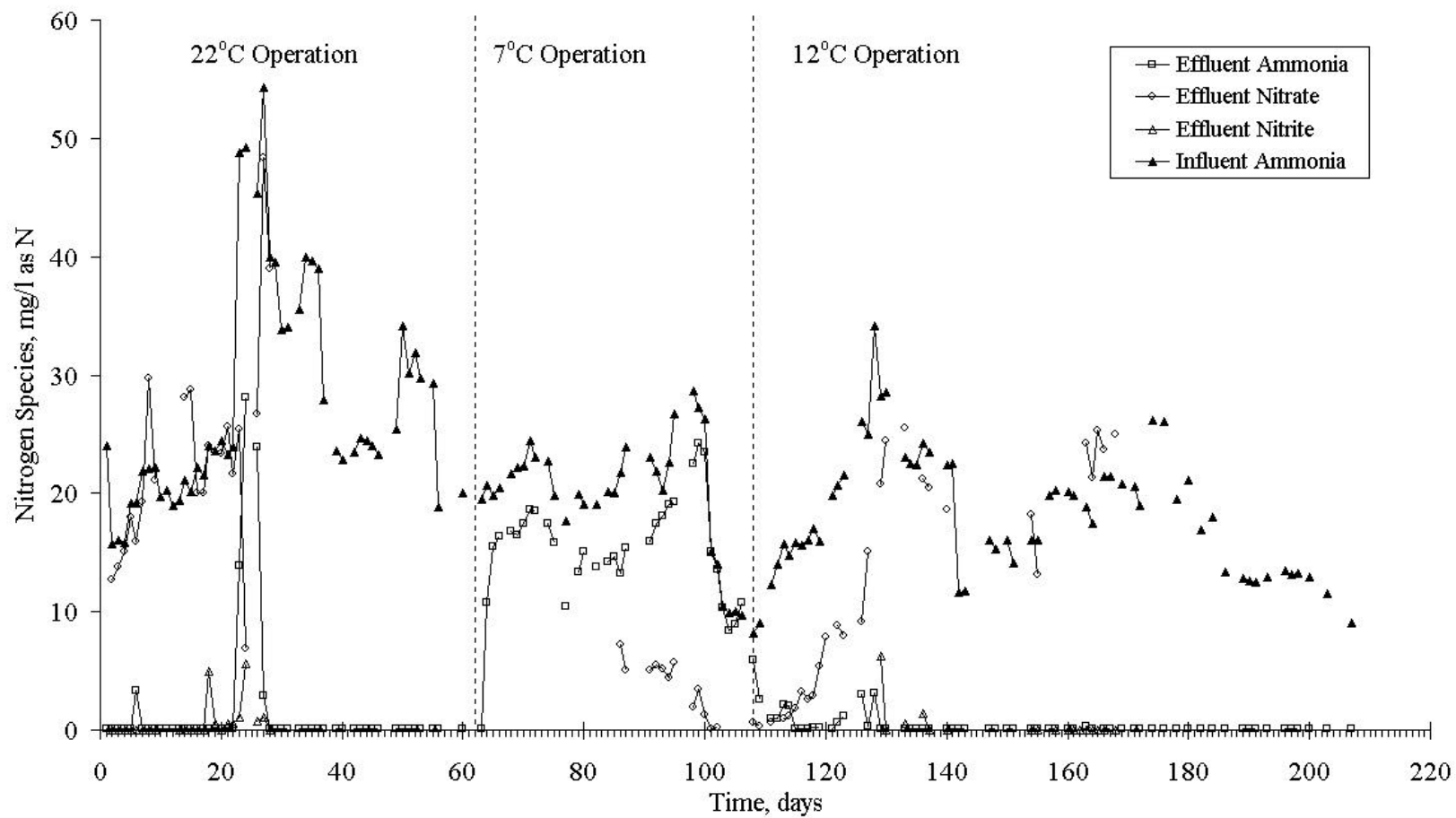


Figure 16. Nitrogen speciation plot for the 10-day SRT indicating the occurrence of complete nitrification.

(beginning on day 23). Based on the period of complete nitrification before the perturbation and the period of complete ammonia removal after, the 10-day SRT system steady state operation was set from day 1 to day 20 and from day 37 to day 60 during 22°C operation. Much like the 20-day SRT system, nitrification performance decreased drastically after the temperature was lowered to 7°C. The average ammonia removal efficiency during 7°C operation was 23.4% (based on ammonia-N removal) indicating poor ammonia-N removal. No steady state was reached at this operational temperature.

Operation at 12°C proved to be successful. The period of acclimation was similar to that of the 20-day SRT reactor. Effluent nitrate concentrations steadily climbed to levels similar to those measured for influent ammonia. Effluent ammonia concentrations returned to levels below detection shortly after the temperature increase. Nitrate data suggests that steady state was not reached until day 133. During the period of nitrifier recovery, nitrite was detected in the effluent (day 129 and day 136). Based on this data, steady state was set between the days of 137 and 207.

Figure 17 presents the nitrogen speciation plot for the 5-day SRT. The average effluent nitrate concentration during the first three weeks of 22°C operation was  $21.0 \pm 3.1$  mg/L-N while the average ammonia concentration was  $27.4 \pm 2.6$  mg/L-N. Nitrite was detected at levels of approximately 1 mg/L as N from day 5 to day 9, days 14 and 15 and days 18 through 21. During the third week, effluent nitrate-N and influent ammonia-N levels were essentially the same indicating complete nitrification. After the perturbation on day 23, effluent ammonia data (below detection limit) suggest that complete nitrification was reestablished. Based on the data available, steady state was set from day 37 to day 60.

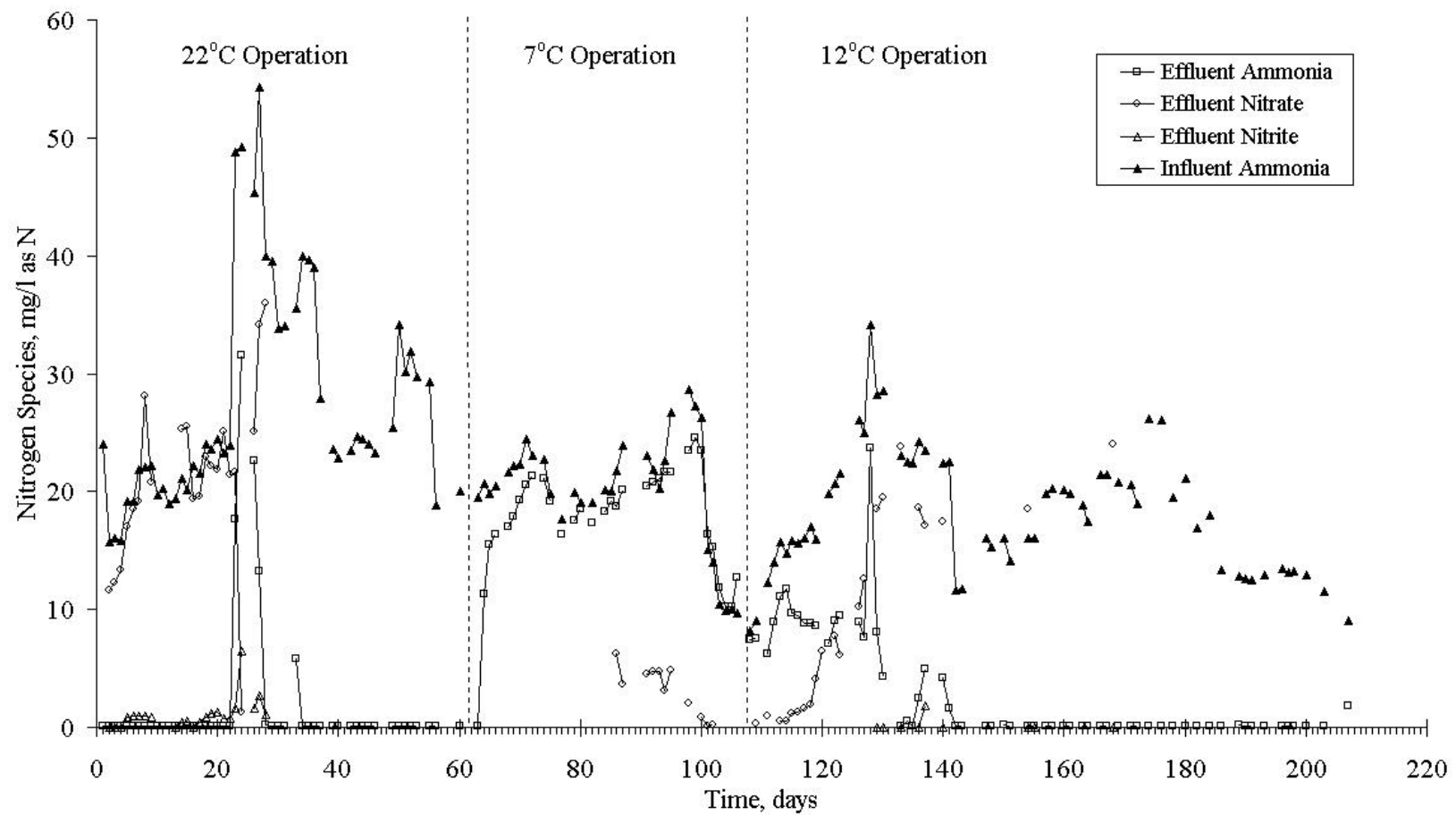


Figure 17. Nitrogen speciation plot for the 5-day SRT indicating the occurrence of complete nitrification.

Nitrification was very poor during operation at 7°C for the 5-day SRT reactor. The average effluent ammonia concentration was  $17.3 \pm 1.7$  mg/L-N while the average influent ammonia concentration was  $19.8 \pm 1.8$  mg/L-N. This level of ammonia removal ( $\approx 2.5$  mg/L-N) could be attributed to cellular synthesis and reproduction. However, based on the nitrate data collected from day 86 to day 102, it appears that a minimal level of nitrification did occur. Nitrate levels dropped below detection levels during the latter part of 7°C operation indicating that nitrification ultimately ceased at this temperature.

Operation at 12°C for the 5-day SRT reactor showed a slower recovery of nitrification than the 10- and 20-day SRT reactors. From day 109 to day 141, the ammonia-N level decreased to below the detection limit (0.1 mg/L-N). During this same time period, effluent nitrate concentrations rose steadily reaching values in close proximity to influent ammonia concentrations. This one month acclimation period illustrated the slow reactivation or reintroduction of nitrifiers under these conditions.

Oleszkiewicz and Berquist (1988) also encountered a gradual population shift in biomass during temperature changes. Gaudy and Gaudy (1980) showed that prolonged low temperature causes both psychrophilic and psychrotrophic organisms to reach an optimum population mix; however, the role of these type of bacteria are not well understood. In this research, complete nitrification was reestablished in the 5-day SRT reactor and steady state for 12°C operation was designated from day 143 to day 203.

Figure 18 presents the nitrogen speciation plot for the 2-day SRT reactor over time. During the initial 22-day operational period at 22°C, the average effluent ammonia concentration in the 2-day SRT reactor was  $3.6 \pm 1.3$  mg/L-N while the average influent

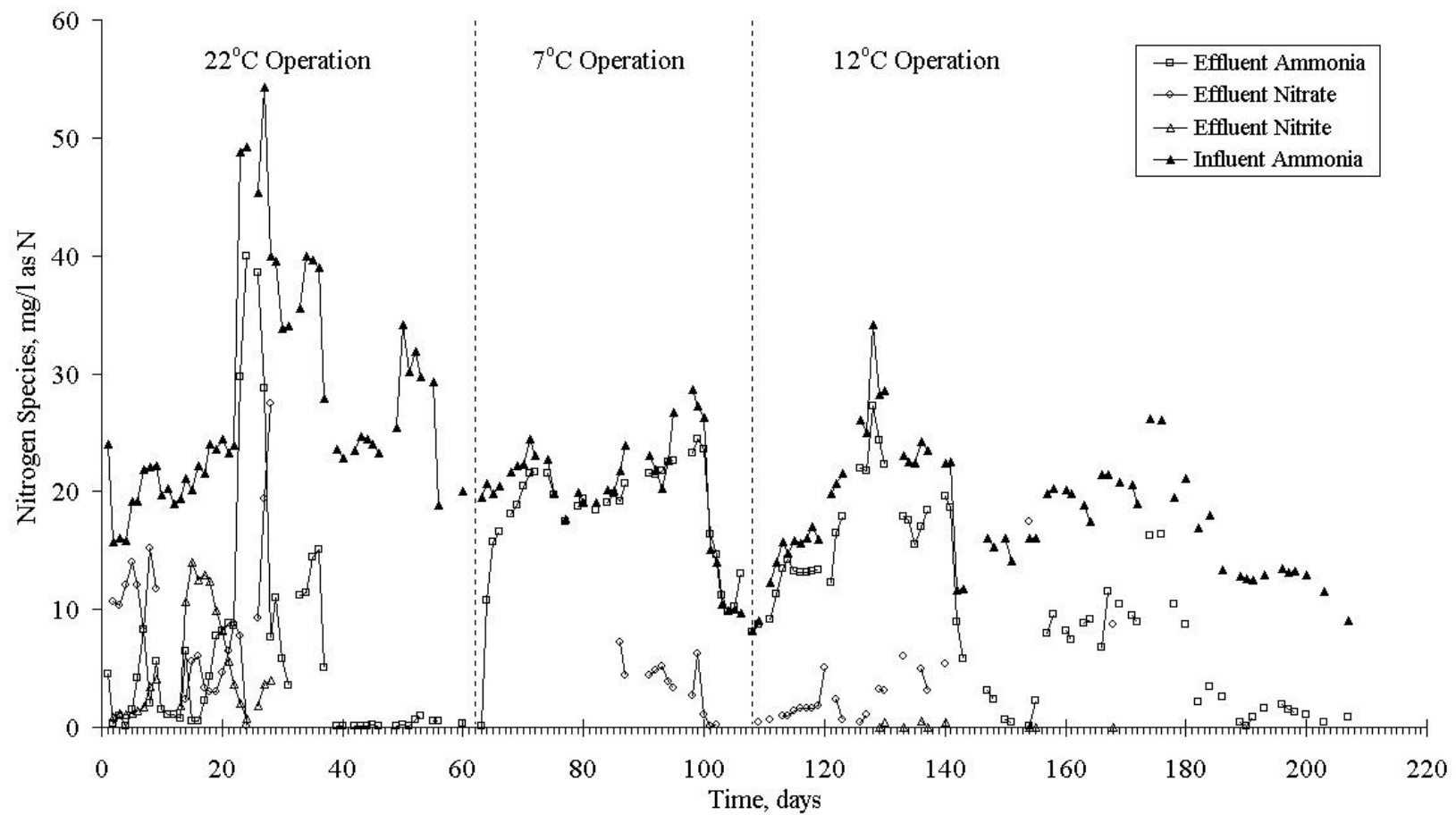


Figure 18. Nitrogen speciation plot for the 2-day SRT indicating the occurrence of complete nitrification.

ammonia concentration was  $20.8 \pm 1.1$  mg/L-N. This represents an average removal efficiency of  $83.8 \pm 5.6$  percent. The average effluent nitrate concentration during the initial 22-day period was  $8.1 \pm 2.0$  mg/L-N; however, the nitrate level decreased during the latter part of this period. The effluent ammonia concentration increased during the same time. Both of these events were preceded by a steady increase of nitrite in the effluent. Nitrite levels increased to 14 mg/L-N in 2 days, then steadily decreased even after the perturbation event had begun. The average nitrite concentration during initial operation was  $5.9 \pm 2.3$  mg/L as N. During the perturbation, the effluent ammonia concentration increased with increasing influent ammonia. A subsequent decrease in effluent ammonia concentration coincided with an increased effluent nitrate concentration. Minor vacillation in the effluent ammonia concentration occurred after the perturbation, therefore steady state for the 2-day SRT system was set from day 41 to day 60 for operation at 22°C.

Operational data at 7°C show that nitrification was poor at this temperature and SRT. The average effluent ammonia concentration was  $17.7 \pm 1.8$  mg/L-N while the average influent ammonia concentration was  $19.8 \pm 1.8$  mg/L-N. Resulting ammonia removal efficiency during 7°C operation was  $8.6 \pm 6.8$  percent. Negative ammonia removal efficiencies were calculated for seven non-consecutive days during 7°C operation. No steady state for nitrification was achieved during 7°C operation.

Operation at 12°C for the 2-day SRT reactor was highly unstable for the majority of time at this temperature. Poor nitrification efficiency persisted until day 154 when effluent ammonia levels dropped below detection limits. However, a large sustained elevation in the effluent ammonia concentration was measured after this date. On day

190, the effluent ammonia concentration returned to levels below the detection limit although an effluent ammonia concentration less than 0.1 mg/L-N was not sustained over time. From day 190 to day 207, a stable effluent ammonia concentration of 1.1 mg/L-N was (on average) measured and this time period was chosen to represent steady state operation.

#### **4.5.3 Alkalinity Observations**

During this bench-scale study, periodic measurements of influent and effluent alkalinity (as  $\text{CaCO}_3$ ) were made (Figure 19) to ensure adequate buffering capacity. As indicated in Equation 1, free hydrogen ions are produced during ammonia oxidation, the first step of nitrification. The potential for pH depression due to acidification from ammonia oxidation poses an operational problem for both treatment of carbonaceous and nitrogenous compounds as it effects both heterotrophic and autotrophic bacterial growth rates (Grady *et al*, 1980). Reactor pH conditions have been found to have a significant effect on nitrification rates (USEPA, 1993).

Theoretical alkalinity consumption during ammonia oxidation can be calculated as 7.14 mg  $\text{CaCO}_3$  / mg  $\text{NH}_4^+$  oxidized. This ratio neglects bacterial synthesis. The average ratio of alkalinity destroyed per ammonia oxidized for each test system is summarized in Table 8. The large variance is due, in part, to the small number of samples taken during the periods of operation.

There is a trend of increasing average alkalinity consumption with increasing SRT during 22°C operation; however, differences are not statistically significant at the 95% confidence level. In general, alkalinity consumption was lower than predicted in reactors that achieved high levels of nitrification.



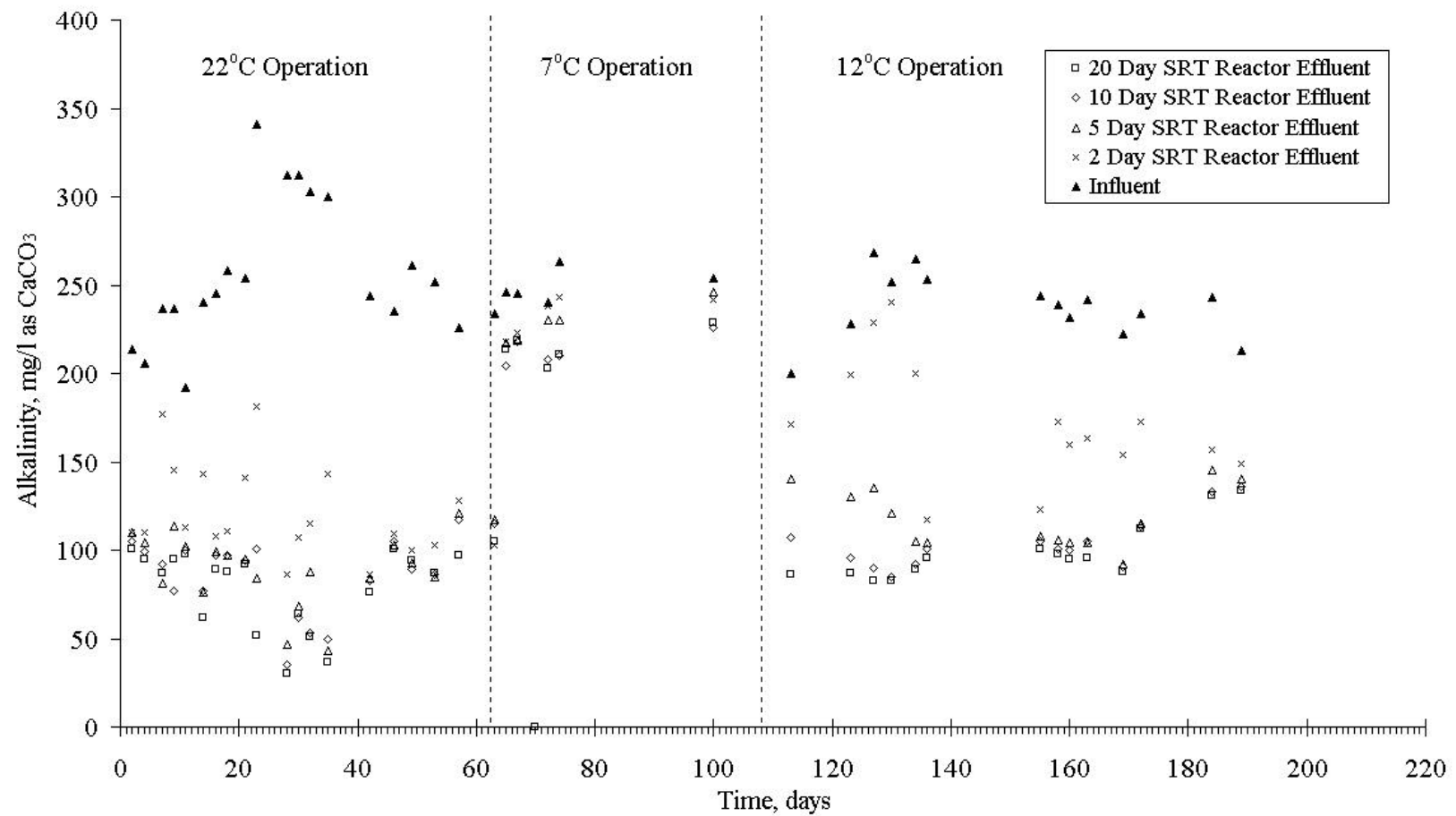


Figure 19. Fluctuations in effluent alkalinity as a result of decreasing temperature throughout this treatability study.

Table 8. Average ratio of alkalinity destroyed per ammonia oxidized expressed as mg  $\text{CaCO}_3$  / mg  $\text{NH}_4^+\text{-N}$ .

Reactor	22°C Operation		7°C Operation		12°C Operation	
	mg $\text{CaCO}_3$ consumed/ mg $\text{NH}_4^+\text{-N}$ oxidized	Std. Dev., mg/L	mg $\text{CaCO}_3$ consumed/ mg $\text{NH}_4^+\text{-N}$ oxidized	Std. Dev., mg/L	mg $\text{CaCO}_3$ consumed/ mg $\text{NH}_4^+\text{-N}$ oxidized	Std. Dev., mg/L
20-day $\theta_c$	6.73	0.40	9.85	1.79	6.97	0.43
10-day $\theta_c$	6.64	0.36	8.57	1.64	6.77	0.40
5-day $\theta_c$	6.58	0.42	8.36	6.15	7.22	1.01
2-day $\theta_c$	6.35	0.54	7.21	5.03	8.58	2.29

Conversely, alkalinity consumption was higher than predicted in reactors that achieved little nitrification (12oC – 2-day SRT and all SRTs at 7oC). During the entire operation of the reactors, the average influent alkalinity concentration was  $248 \pm 10$  mg/L as  $\text{CaCO}_3$ . The average effluent alkalinity for the 20-, 10-, 5- and 2-day SRT reactors was  $104 \pm 15$ ,  $109 \pm 14$ ,  $118 \pm 15$  and  $154 \pm 15$  mg/L as  $\text{CaCO}_3$ , respectively. As indicated by these values, sufficient alkalinity was maintained during operation. However, during the perturbation event, an alkalinity value of 30 mg/L (as  $\text{CaCO}_3$ ) was measured in the 20-day SRT reactor. This depression was due to the increase in ammonia loading on the system, and it was noted that the corresponding reactor pH was 6.62. Low pH values were recorded during this time period; however, these levels did not persist after wastewater strength returned to normal.

## **4.6 Examination of Full Scale Simulation Using the Bench Scale**

### **Reactor / Clarifier System**

A unique feature of this study was that actual settled municipal wastewater was used in order to more accurately simulate the local full scale activated sludge process. The reactor / clarifier system was also seeded with biomass collected from the local full scale WWTP activated sludge reactor. An analysis of treatment performance between the two systems was required to identify areas in which the bench scale system sufficiently simulated the full scale system.

Inherent differences between the bench and full scale facilities did exist. For instance, the hydraulic retention time (HRT) for the full scale facility was approximately 3.4 hours whereas an HRT of the bench scale system was approximately 8.6 hours. Hydraulic and mixing characteristics between the two systems were theoretically different and a discussion of similitude was not approached for this study. However, a comparison of carbon treatment, sludge settlability, and ammonia removal between the two systems was made to identify successful treatment simulation.

Figure 20 shows the operational SRT for the full scale system during this study. The average SRT for the full scale activated sludge system was approximately 3 days. For comparative purposes, the 5-day SRT treatment data was chosen to represent the bench scale system. Figure 21 illustrates the change in operational temperature for the full scale facility during this bench scale study. Full scale temperature steadily declined from an initial temperature of approximately 23°C to temperatures as low as 4°C. Since

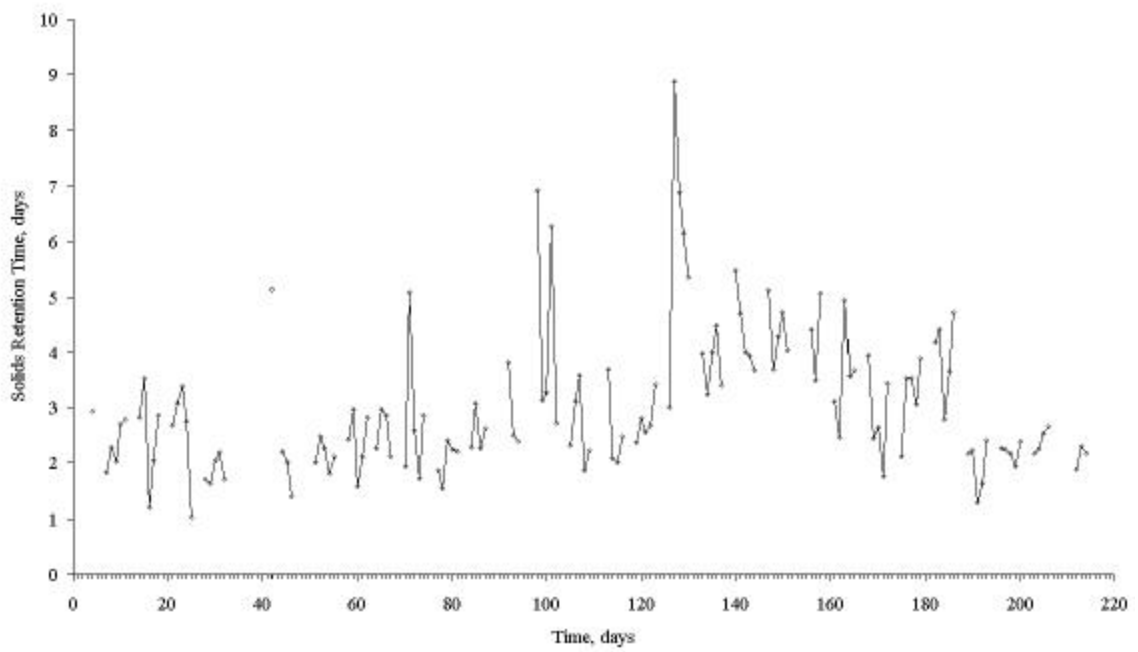


Figure 20. A plot of operational SRT for KUBs Kuwahee WWTP during this study.

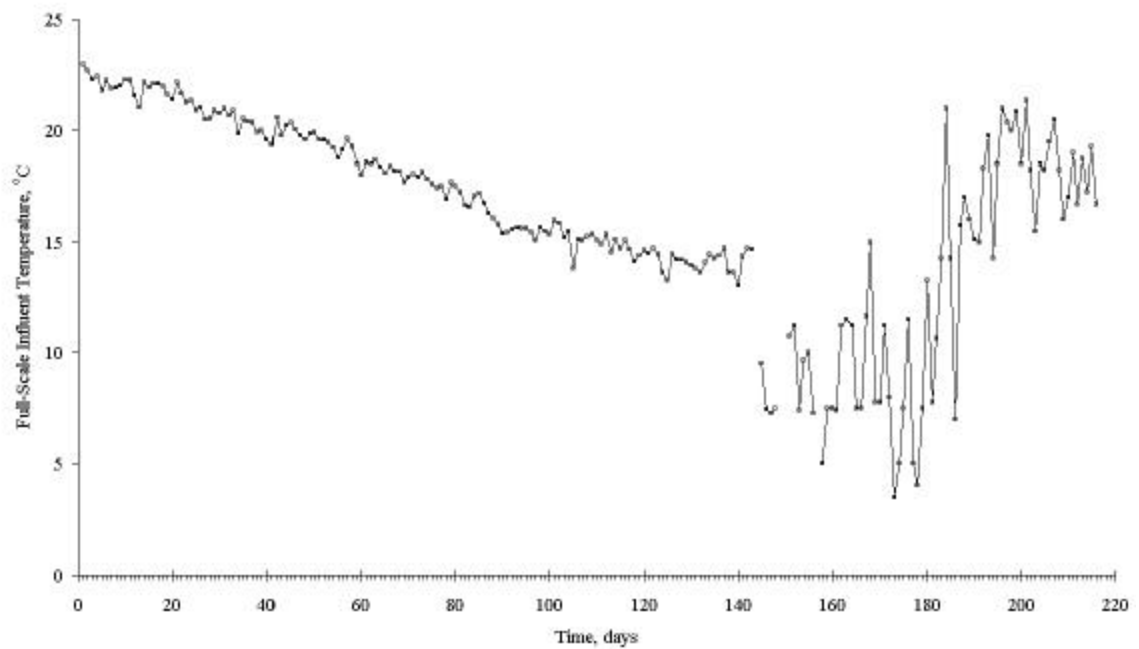


Figure 21. A plot of influent temperature for KUBs Kuwahee WWTP during this study.

this study was performed between the dates of September 28, 1999 to April 27, 2000, the temperature drop in Figure 21 encompassed wintertime conditions.

Figures 22 and Figure 23 show the carbon treatment data for both the full and bench scale (5-day SRT) systems. Due, in part, to the inherent differences in measurement between  $BOD_5$  and  $COD_T$ , influent values were quite scattered, unexpectedly. The increase in wastewater strength observed during the initial perturbation in the bench scale system was not as pronounced in the full scale data. This could be attributed to a volume effect where a greater amount of equalization of the high strength wastewater occurred in the full scale system. As can be seen in Figure 22, influent  $BOD_5$  for the full scale system was, at times, much greater than values observed during the perturbation. In addition, other periods of high strength influent wastewater measured at the full scale facility were not observed in the bench scale system. Since wastewater was collected every 3-4 days, many influent high strength events were not captured in the bench scale operation. Figure 23 shows that effluent  $COD_T$  values were much greater in the bench scale system than the full scale effluent  $BOD_5$  values. This was, in part, attributed to the suspended solids which were included in the effluent  $COD_T$  measurements. As can be seen in Figure 23, the full scale facility produced an effluent with very low biochemical oxygen demand. Residual oxygen demand is of main concern for WWTP operation, due to detrimental effects of chlorination when oxidizable organic matter is present as well as effects of high residual oxygen demand wastewater on receiving streams. Despite periods of clear, well treated effluent, the bench scale system was more sensitive to process changes causing higher, more variable effluent  $COD_T$  values.

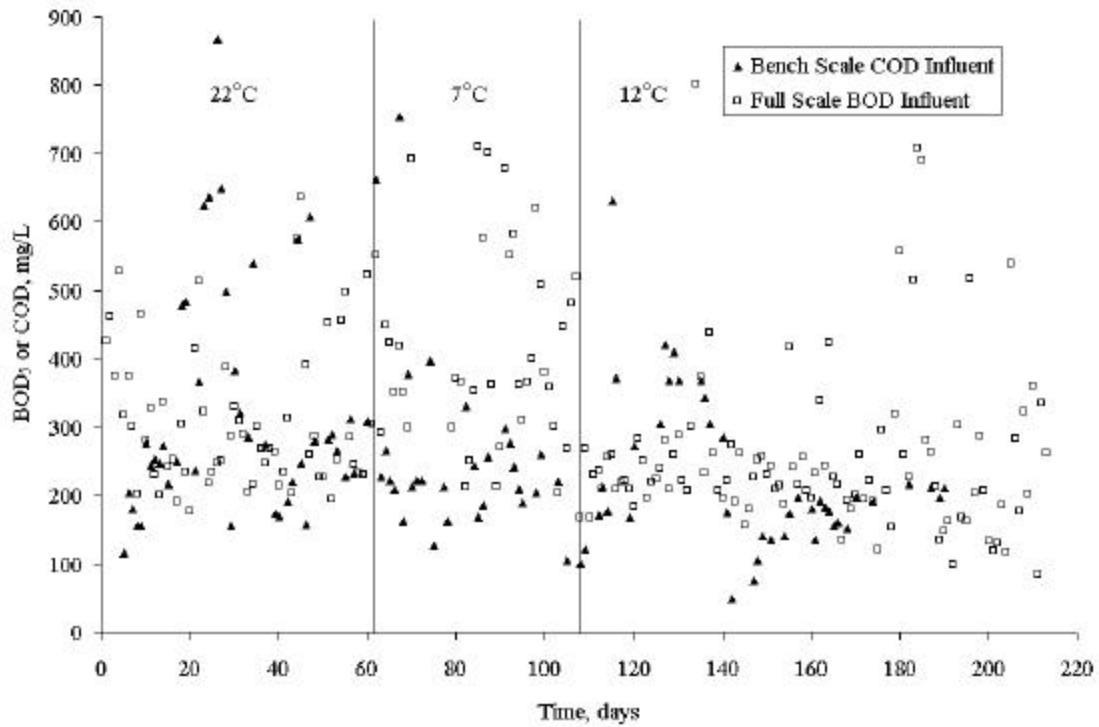


Figure 22. A comparison of full and bench scale (5-day SRT) influent carbon data during this study.

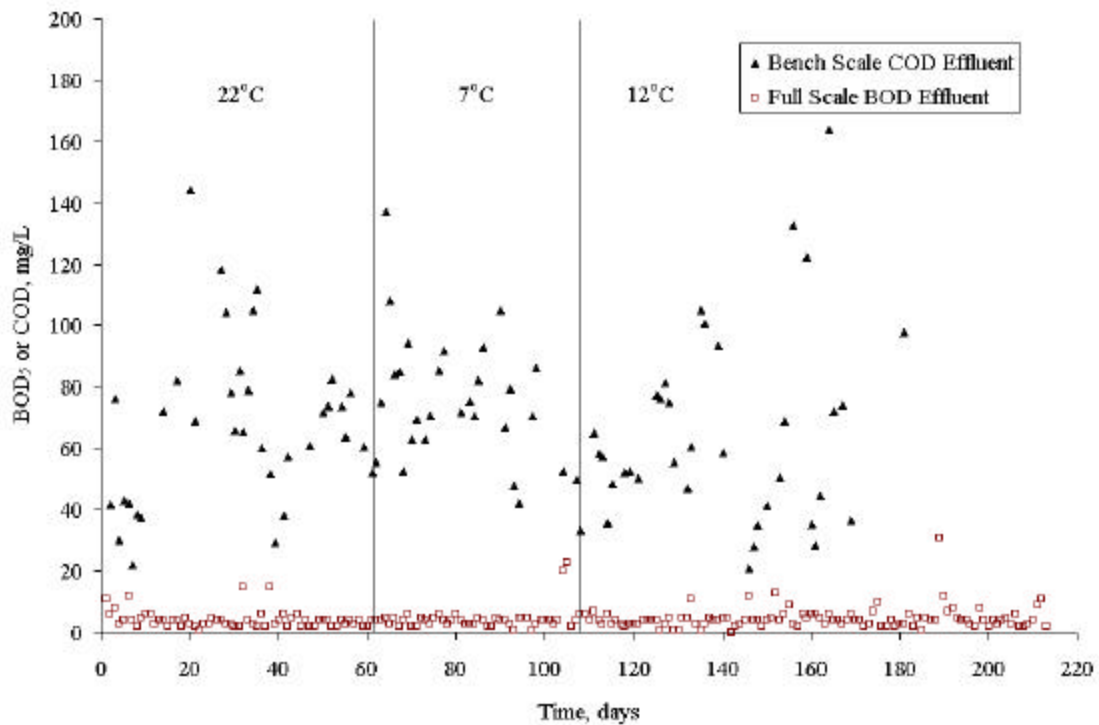


Figure 23. A comparison of full and bench scale (5-day SRT) effluent carbon data during this study.

Overall, it was difficult to compare full and bench scale carbon treatment data based on differences in carbon measurement, differences in flow, variation in wastewater strength, and differing operational (SRT) and environmental (temperature) parameters between the two systems.

Figure 24 represents the MLSS concentration maintained in both the full and bench scale (5-day SRT) systems during this study. A large difference in MLSS concentration between the two systems was observed for the entire duration of this study. Average MLSS concentrations for the full scale system for the defined temperature intervals of 22, 12 and 7°C were 3858, 3100 and 2950 mg/L, respectively. The average MLSS values for the bench scale system during the established temperature intervals of 22, 12 and 7°C were 1624, 775, 1269 mg/L, respectively. The average MLSS values were markedly different between the two systems. Although an MLSS peak during operation between days 20 through 40 was noted in both systems.

Figure 25 illustrates changes in SVI for both the full and bench scale (5-day SRT) systems during this study. Several major vacillations in SVI were measured in the bench scale system during the first 2 months of operation; however, SVI values ultimately stabilized at levels comparable to the full scale facility. Biomass acclimation and operational temperature were identified as playing a role in the achievement of excellent SVIs in the bench scale system by the end of this study.

Figure 26 shows the F/M ratios for both the full and bench scale (5-day SRT) systems during this study. F/M ratio for each system was based on the means by which carbon substrate was measured.

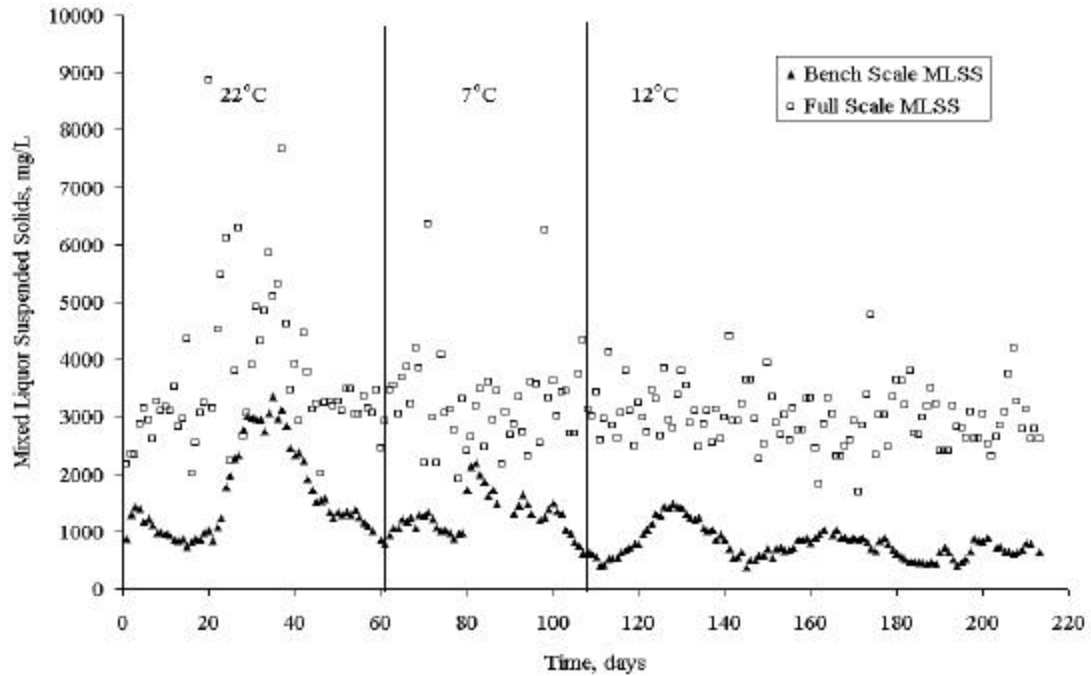


Figure 24. A comparison of Mixed Liquor Suspended Solids for both the full and bench scale systems during this study.

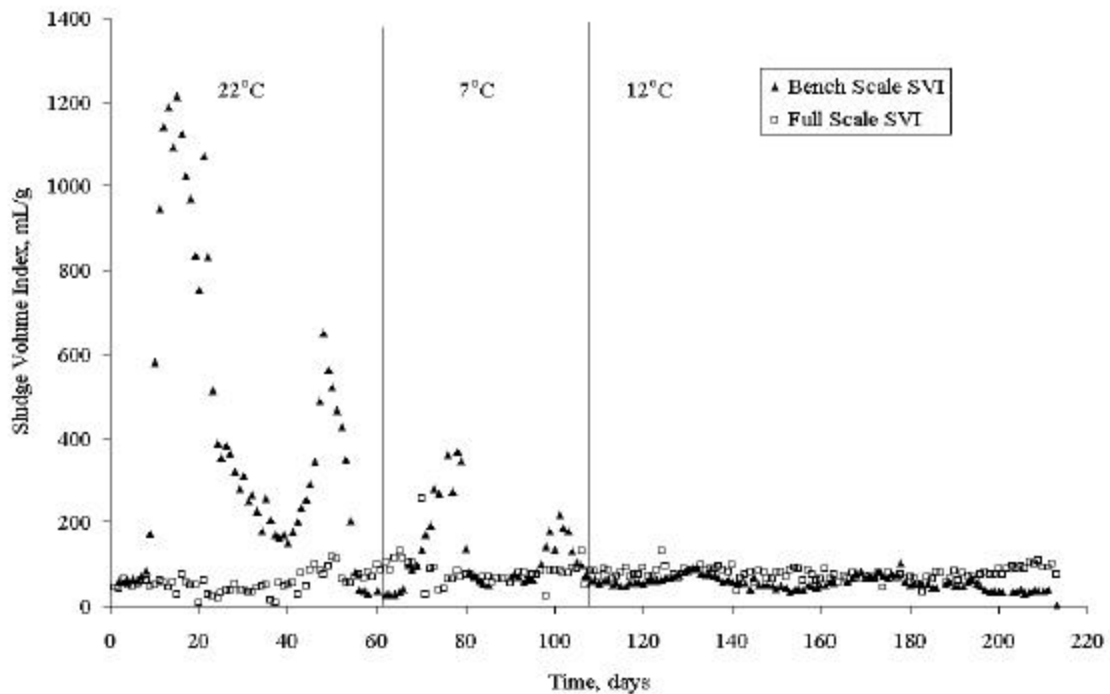


Figure 25. A plot of SVI for the full and bench scale (5-day SRT) systems during this study.



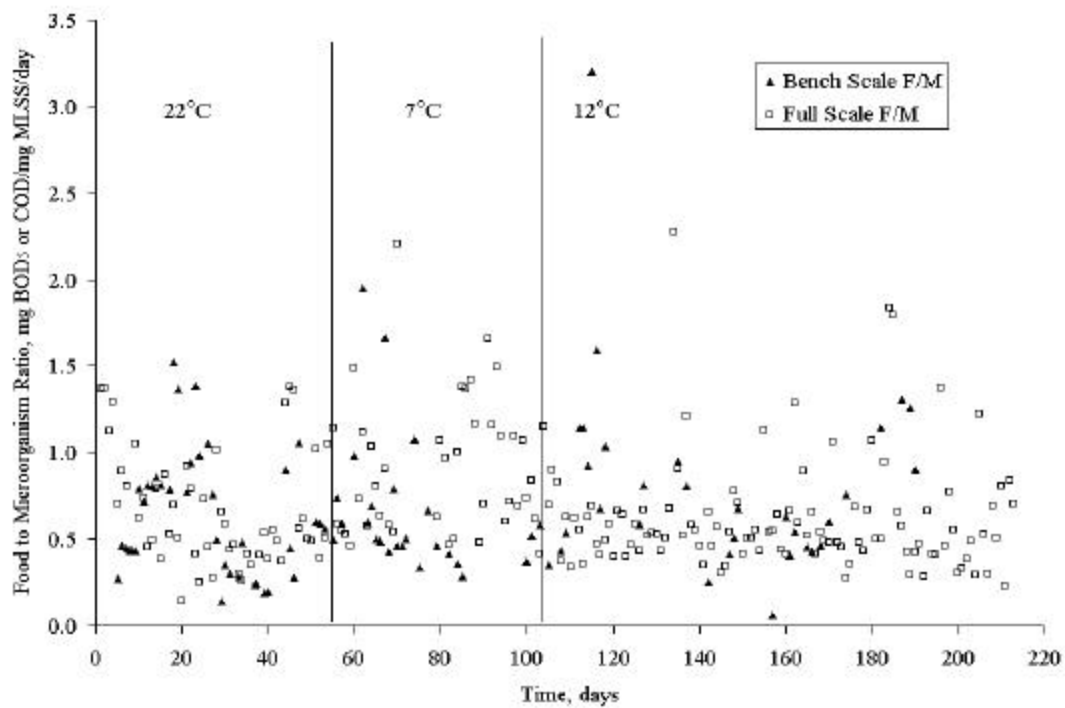


Figure 26. A plot of F/M ratios for the full and bench scale (5-day SRT) systems during this study.

It was difficult to compare F/M ratios between the two systems, as was the case for carbon treatment data, due to the differences in methods used to assess influent carbon concentrations. Although F/M ratios for the bench and full scale systems do coincide over certain time frames, there is little general agreement in values over the entire time course of the study.

Figures 27 and 28 show the influent and effluent ammonia data, respectively, for both the full and bench scale (5-day SRT) systems over the duration of this study. The full and bench scale systems had somewhat similar influent ammonia concentrations; however, the bench scale system ammonia concentration was consistently higher. A comparison of average values for the temperature regimes of 22, 12 and 7°C for the full

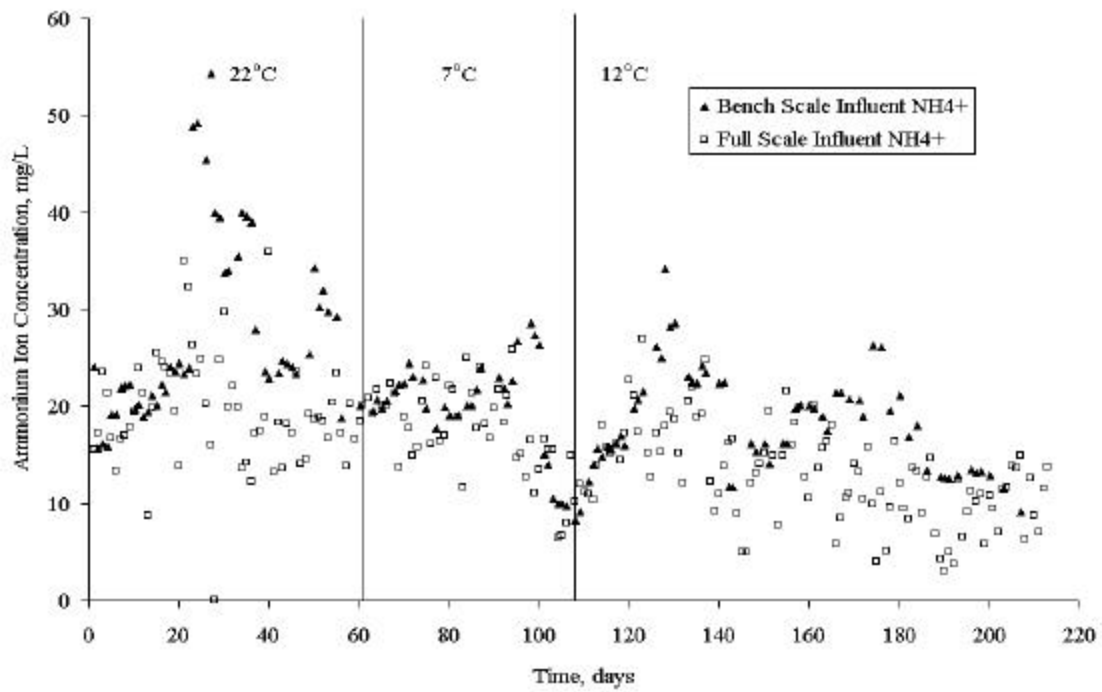


Figure 27. A plot of ammonia treatment data for both the full and bench scale (5-day SRT) systems for this study.

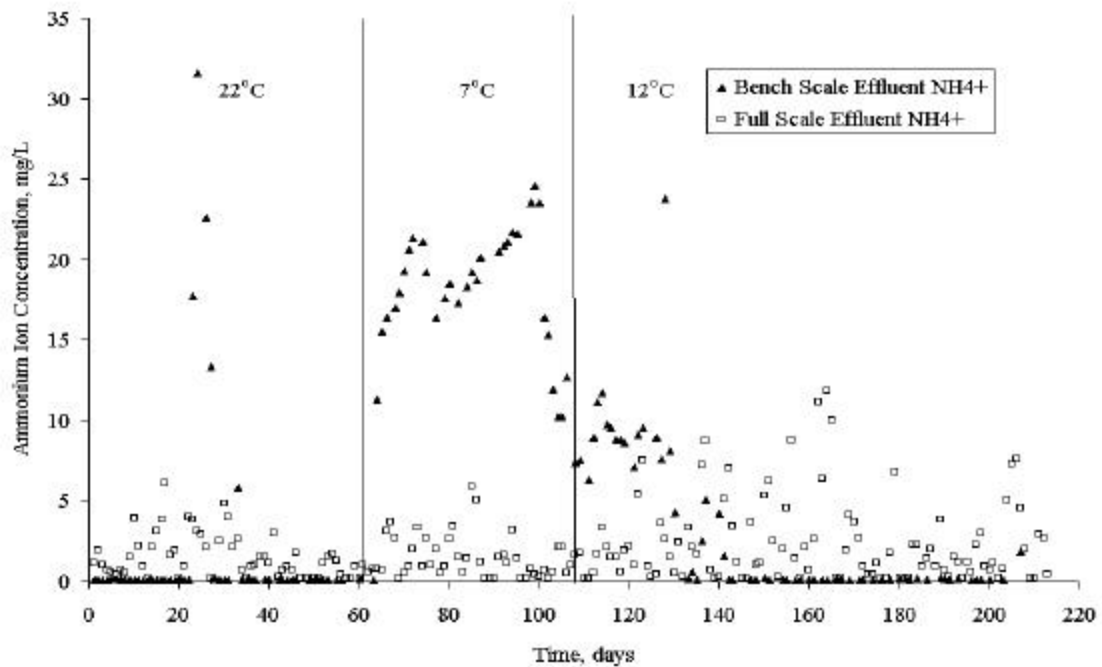


Figure 28. A plot of ammonia treatment data for both the full and bench scale (5-day SRT) systems for this study.

and bench scale systems were 19.9 and 27.4, 16.6 and 18.2, and 12.8 and 19.8 mg/L, respectively. Conversion of organic nitrogen to ammonia through the process of ammonification and time of day at which the settled municipal wastewater was collected were thought to have been the main reasons for this consistent difference in influent ammonia concentrations between the two systems. Figure 28 shows the effluent ammonia concentrations for both systems. The substantial residual ammonia in the full scale effluent could have been due to the low operational SRT (approximately 3 days on average) maintained for the full scale facility. Operationally, a small amount of ammonia is acceptable for full scale operation based on permitted effluent discharges. In comparison, the bench scale system produced effluent with lower ammonia concentrations than the full scale facility. The discrepancy in ammonia treatment performance could have been due to a higher operating SRT for the bench scale system or a higher concentration of ammonia oxidizing bacteria within the reactor.

Overall, from the data presented in this section, the bench scale reactor / clarifier system had difficulty in simulating the local full scale WWTP. Similarities did exist between the two systems; however, differences in system operation, waste collection, measurement of carbon substrate, bench scale biomass acclimation and other environmental parameters were thought to have played a role in lack of a successful comparison between the two systems.

The previous graphs comparing full and bench scale (5 day SRT) systems provided collation of operating data for the time frame of this study. To assess the performance of both systems based on the major environmental parameter, temperature, periods of similar operational temperature were compared. The average full scale

process temperature from day 1 to day 37 was found to be 21.5°C; whereas, the average full scale process temperature from day 111 to day 143 was found to be 14.3°C and finally from day 145 to day 181 the average full scale process temperature was determined to be 8.6°C. These periods were chosen to approximate the temperatures imposed (22, 12 and 7°C) on the bench scale system. Average treatment data for the full and bench scale systems during these periods of similar temperature are presented here as Table 9.

For the full scale system, the average values for carbon treatment data presented in Table 9 showed little variation in treatment performance with respect to temperature. For the 22, 12 and 7°C full scale temperature approximations, the average effluent BOD<sub>5</sub> with accompanying standard deviations at a 95% confidence interval in milligrams per liter were  $4.4 \pm 2.9$ ,  $3.7 \pm 2.0$ , and  $4.8 \pm 2.6$ , respectively. Average effluent BOD<sub>5</sub> values for different temperature periods did not vary significantly. The most efficient carbon treatment occurred at 14.3°C (98.56%) followed by 21.6°C (98.6%) with treatment at 8.6°C (98.0%) exhibiting the lowest BOD<sub>5</sub> removal. Despite a decline in average influent BOD<sub>5</sub> with respect to temperature, effluent BOD<sub>5</sub> values remained low and subsequent treatment performance continued to be relatively constant independent of temperature. Due to different means of measuring carbon substrate, bench scale system did not successfully simulate the full scale system with respect to carbon treatment performance; even though, a similar lack of temperature response to carbon treatment was observed in both systems. However, it was believed that a more accurate analysis of simulation of full scale carbon treatment could have been possible if BOD<sub>5</sub> data was measured in the bench scale study.

Table 9. Average carbon and nitrogen values for the full scale and bench scale systems during periods of similar operating temperature.

System, Avg. Temp.	Days of Operation	Average Influent COD/BOD <sub>5</sub>	Average Effluent COD/BOD <sub>5</sub>	Average Influent NH <sub>4</sub> <sup>+</sup>	Average Effluent NH <sub>4</sub> <sup>+</sup>
Full Scale, 21.5°C	37	304.5 ± 91.4	4.4 ± 2.9	19.9 ± 6.5	1.9 ± 1.5
Bench Scale, 22°C	61	323.3 ± 169.9	67.8 ± 26.9	27.4 ± 9.4	1.9 ± 6.1
Full Scale, 14.3°C	33	261.1 ± 110.3	3.7 ± 2.0	16.6 ± 4.1	2.5 ± 2.4
Bench Scale, 12°C	105	226.1 ± 113.3	64.9 ± 31.6	18.2 ± 5.2	3.1 ± 4.7
Full Scale, 8.6°C	37	239.8 ± 83.5	4.8 ± 2.6	12.8 ± 4.6	2.9 ± 3.3
Bench Scale, 7°C	47	257.4 ± 140.1	75.1 ± 20.8	19.8 ± 5.2	17.3 ± 5.1

A comparison of ammonia treatment data between bench and full scale systems at similar temperatures revealed a more distinct effect of temperature on system performance. For temperatures of 22, 12 and 7°C, the full scale system average effluent ammonia concentrations with corresponding standard deviations at a 95% confidence interval in milligrams nitrogen per liter were  $1.9 \pm 1.5$ ,  $2.5 \pm 2.4$  and  $2.9 \pm 3.3$ , respectively. Despite the close proximity of average effluent ammonia concentrations, an increase in effluent ammonia was observed with decreasing temperature. This direct relationship was expected based on a review of literature (Section 2.4) and was also observed in the bench scale system. At 7°C for the bench scale system, a dramatic loss of ammonia removal attributed to low temperature was evidenced by the elevated average effluent ammonia concentration of  $17.3 \pm 5.1$  mg/L compared to that of  $2.9 \pm 3.3$  mg/L for the full scale system. The bench scale system underwent an almost total loss of ammonia removal at 7°C whereas the full scale system only experienced a slight reduction in ammonia removal during the average temperature period of 8.6°C.

The difference in operational temperature between the two systems (7°C versus 8.6°C) could have, in part, caused some difference in operation between the two systems. Also, the SRT observed for the bench scale system was fixed (5 days) while the SRT for the full scale system was elevated during colder temperatures. This elevation of full scale SRT began during the period considered for 12°C operation.

For 22°C, the average effluent ammonia concentrations with standard deviations at a 95% confidence interval for the full and bench scale systems were  $1.9 \pm 1.5$  and  $1.9 \pm 6.1$  mg/L, respectively. For 12°C the average effluent ammonia concentration with standard deviation at a 95% confidence interval were  $2.5 \pm 2.4$  and  $3.1 \pm 4.7$  mg/L, respectively. At 22 and 12°C the bench scale system had higher ammonia removal efficiencies than the full scale system while experiencing higher, more variable ammonia loading. There was, however, an indirect relationship between decreasing temperature and increasing average effluent ammonia concentration. This temperature dependant relationship was observed in both the full and bench scale systems to varying degrees. Full scale treatment performance for ammonia removal was not effected by temperature as the bench scale system. Even though the same indirect relationship between temperature and effluent ammonia concentration was observed in both reactors, this effect was found to occur at varying degrees. Temperature inhibition in the full scale system was less dramatic; whereas, the same effect was quite dramatic in the bench scale system. The bench scale system consistently produced an effluent with lower ammonia concentrations. Overall, the bench scale simulation of full scale ammonia oxidation was quite good despite a multitude of different operational and environmental variables.

## Chapter 5.0

### Summary and Conclusions

#### 5.1 Evaluation of Bench Scale Reactor Design and Operation

This bench-scale, recycle system was sufficient in meeting expectations during the period of operation. Tight control of SRT was achieved through daily wastage of solids. The external clarifier performed well after several modifications, achieving effluent suspended solids levels below 30 mg/L on a consistent basis. This attribute was due, in part, to the scraper arm portion of the external clarifier which facilitated solids movement and concentration, allowing for proper solids recycle. Average effluent suspended solids concentrations during the entire length of operation were  $24.5 \pm 2.8$ ,  $21.6 \pm 3.2$ ,  $22.7 \pm 2.4$ ,  $20.4 \pm 2.7$  mg/L for the 20-, 10-, 5- and 2-day SRT reactors, respectively at a 95 % confidence interval. SVI values were poor initially; however, settling improved upon temperature change to 12°C. During 12°C operation, average SVI values were 57, 65, 55 and 69 L/mg, indicating conditioned, well settling biosolids.

Reactor temperature was maintained through set point controls for the constant temperature room where the bench-scale system was operated. Periodic checks of the reactor temperature confirmed consistent reactor temperature, however the temperature did maintain approximately 1.5°C above the set point for all temperatures studied.

Kuwahee wastewater used during this experiment had sufficient alkalinity to buffer pH in the mixed liquor. Concentrations of > 80 mg/L were typically measured in the effluent except during periods of high-strength influent wastewater. Alkalinity is

important in system pH, which can effect the growth of bacteria. Fortunately sufficient alkalinity was present and pH control was not required.

## **5.2 Evaluation of Carbon Treatment Performance**

An objective of this treatability study was to assess the impact of temperature and SRT on carbon removal efficiency. At 22°C operation, the average effluent COD increased with increasing SRT. Statistical analysis indicated that treatment performance was a function of SRT at this temperature. However, no distinct relationship between SRT and COD removal was noted within the 7°C or 12°C operational temperatures. However, statistical analysis showed a significant difference in the average effluent COD values between the 22°C and 7°C operational temperatures. These findings indicate that effective carbon treatment can be accomplished at temperatures above 12°C.

## **5.3 Examination of Nitrification Performance**

An additional objective of this study was to determine the effects of temperature and SRT on nitrification. Nitrification was successfully achieved for this bench-scale system at temperatures as low as 12°C and at an SRT of 5 days or greater. The 20, 10 and 5-day SRT reactors successfully achieved complete ammonia removal, however the 2-day SRT was not sufficient to consistently achieve complete nitrification at any temperature studied. Therefore a minimum SRT of 5 days is deemed necessary for effective nitrification.



## **5.4 Evaluation of Bench Scale Simulation of Full Scale System**

An objective of this study was to evaluate the efficacy of the bench scale system in simulating of the local full scale WWTP. A comparison of the carbon data revealed a consistent level of treatment for both systems. However, effluent  $\text{BOD}_5$  for the full scale system was very low compared to effluent  $\text{COD}_T$  values obtained from the bench scale system. The bench scale system was found not to successfully simulate the full scale system due, in part, to differences in carbon analysis. A comparison of nitrogen treatment data for both systems bared similar results despite different operational characteristics, such as temperature, variations in wastewater flow and operational SRT. Nitrogen treatment in the bench scale system was more efficient than in the full scale system. This was thought to be due to difference in system operation and other factors (SRT, temperature, waste collection and storage) inherent in both systems. An analysis of the settling characteristics for both systems showed that once the biomass had acclimated in the bench scale system, settling characteristics for both systems were very similar. F/M ratios were highly variable in the two treatment systems. From this comparison of the two systems, the bench scale system designed and fabricated for this study did not simulate the full scale facility as well as expected, in spite of some operational similarities.

## 5.5 Conclusions

The following conclusions can be drawn from this investigation.

1. The bench scale reactor / clarifier system proved to operate sufficiently allowing for tight control of SRT, producing clear, well-treated effluent by the end of the study (12°C operation with an average influent COD<sub>T</sub> of 213 mg/L).
2. Statistical analysis indicated that treatment performance was a function of SRT at 22°C operation supported by the fact that the average effluent COD<sub>T</sub> decreased with increasing SRT. Also, no distinct relationship between SRT and COD<sub>T</sub> removal was derived due to lack of conclusive data gathered at 7°C and the indistinct relationship between SRT and COD<sub>T</sub> treatment performance at 12°C.
3. The bench scale reactor / clarifier system successfully simulated nitrification (above a temperature of 12°C) as well as settling characteristics for the local full scale WWTP activated sludge reactor after appropriate acclimation of the bench scale system; however, carbon treatment was not observed to successfully simulate the local full scale WWTP system due to inherent differences in variations in wastewater strength and means of measuring carbon substrate (BOD<sub>5</sub> v COD<sub>T</sub>).
4. Nitrification was consistently and successfully achieved for this bench-scale system at temperatures as low as 12°C and at a SRT of 5 days or greater.
5. Based on the data presented in this study, a SRT of 5 days and a process temperature of 12°C are observed to achieve effective carbon treatment, biosolids settling, and nitrification.

## References

**Anon** (1974) *Process design Techniques for Industrial Waste Treatment*, pp. 215-232.

Aware, Inc., Enviro. Press Nashville, Tenn.

**APHA** (1998) *Standard Methods for the Examination of Water and Wastewater*, 20<sup>th</sup> ed.

United Book Press, Inc., Baltimore, Maryland.

**Arrhenius S.** (1889). "Über die Reaktionsgeschwindigkeit bei der Inversion von Rohrzucker durch Säuren," *Zeitschrift Fur Physikalische Chemic*, **4**, 226-248.

**ASCE Manual and Report on Engineering Practice No. 76.** (1992) *Design of Municipal Wastewater Treatment Plants Volume I: Chapters 1-12. WEF Manual of Practice No. 8.*

**Aziz J.A. and Tebbutt T.H.Y.** (1979) Significance of COD, BOD and TOC correlations in kinetic models of biological oxidation. *Water Research* **14**, p319-324.

**Barnes D. and Bliss P.J.** (1983) *Biological Control of Nitrogen in Wastewater Treatment*. University Press, Cambridge.

**Barritt N.W.** (1933) The nitrification process in soils and biological filters. *Ann. Appl. Biol.* **20**, 165-184.

**Bock E., Koops H.P.** (1992). The genus *Nitrobacter* and related genera. *The prokaryotes* 2<sup>nd</sup> ed. Springer-Verlag, New York, N.Y. 2302-2309.

**Deppe K. & Engel H.** (1960) Untersuchungen über die Temperatureabhängigkeit der Nitritbildung durch *Nitrobacter winogradskyi* Buch. Bei ungehemmtem und gehemmttem Wachstum. *Zentralbl. Bakteriologie. Parasitenkd. Infek. Hyg. Abt. II* **113**, 561-568.

**Downing A.L., and Knowles G.** (1966) Population dynamics in biological treatment plants. *Presented at the 3<sup>rd</sup> Conference of the IAWPR, Munich.*

- Downing A.L., and Hopwood A.P.** (1964) Some observations on the kinetics of nitrifying activated sludge plants. *Schweiz. Zeitsch f Hydrol.*, **26**, 2, p 271.
- Department of Scientific and Industrial Research** (1964) Water Pollution Research, 1963. *HMSO, London*.
- Eckenfelder W.W., Grau P.D.** (1998) Activated Sludge Process Design and Control: Theory and Practice, Water Quality Management Library Vol 1. 2<sup>nd</sup> ed. *Technomic Publishing Co. Inc.*
- Ekama G. A., and Marais G.v.R.** (1984) Nitrification. *Theory, Design and Operation of Nutrient Removal Activated Sludge Processes. Water Research Commission, Chap. 5.*
- El-Rehaili A.** (1994) Implications of activated sludge kinetics based on total or soluble BOD, COD and TOC. *Environmental Technology* **15**, p1131-1172.
- Ford D.L.** (1980) Comprehensive analysis of nitrification of chemical processing wastewaters. *Journal WPCF*, **52**, 11 p 2726-2746.
- Fuchs A., Staudinger G.** (1999) Characterising the clarification of the supernatant of activated sludges. *Water Res.* **33**, 11 p2527-2534.
- Gaudy A.F. and Kincannon D.F.** (1977) Comparing design models for activated sludge. *Water Sew. Works*, **123**, 66.
- Grady C.P.L., Lim H.C.** (1980) *Biological Wastewater Treatment: Theory and Applications*. Marcel Dekker, Inc.
- Grady C.P.L., Smets B.F., Barbeau D.S.** (1996) Variability in kinetic parameter estimates: a review of possible causes and a proposed terminology. *Water Res.* **30**, 742.

**Gromiec M., Valve M. and Liponkoski M.** (1982) *Nutrients removal from wastewaters by the single sludge systems*. Technical Research Centre of Finland, Research Reports 126p.

**Gujer W. and Jenkins D.** (1974) The contact stabilization process-oxygen and nitrogen mass balances. *University of California, Sanitary Engineering Research Lab, SERL Report 74-2*.

**Haug T.T. and McCarty P.L.** (1971) Nitrification with the submerged filter. *Report prepared by the Department of Civil Engineering, Stanford University for the Environmental Protection Agency, Research Grant No. 17010 EPM*.

**Henze, M., Harremoes P., LaCour Jansen J., and Arvin E.** (1995) *Wastewater Treatment: Biological and Chemical Processes*. Springer, Heidelberg.

**IAWPRC Task Group on Mathematical Modeling for Design and Operation of Biological Wastewater Treatment**, "Final report: Activated sludge model." *IAWPRC Sci. Tech. Rep. No. 1*.

**Jones R.D., Morita R., Koops H.P., and Watson S.** (1998). A new marine ammonium-oxidizing bacterium, *Nitrosomonas cryotolerans* sp. Nov. *Can. J. Microbiol.* **3**, 1122-1128.

**Jones J.B. and Dugan R.E.** (1996) *Engineering Thermodynamics*. Prentice Hall, Inc. Juretschko S., Timmermann G., Schmid M., Schileifer K.H., Pommerening-Roser A.,

**Koops H.P., and Wagner M.** (1998) Combined molecular and conventional analyses of nitrifying bacterium diversity in activated sludge: *Nitrosococcus mobilis* and *Nitrospira*-like bacteria as dominant populations. *App. Environ. Microbiol.* **64**, 3042-3051.

- Knowles G., Downing A.L., and Barrett M.J.** (1965) Determination of kinetic constants for nitrifying bacteria in mixed culture, with the aid of an electronic computer. *J. Gen. Microbiology*, **38**, 263.
- Koch G., Kuhni M., Gujer W., and Siegrist H.** (2000) Calibration and validation of activated sludge model no. 3 for Swiss municipal wastewater. *Wat. Res.* **34**, 14, 3580-3590.
- Krishna C., Van Loosdrecht M.C.M.** (1999) Effect of temperature on storage of polymers and settleability of activated sludge. *Water Res.* **33**, 10, p2374-2382.
- Laudelout H.** (1978) Mathematical modeling of biological ammonium oxidation. In *Microbiology – 1978* (Edited by Schlessinger D.) Am. Soc. Microbiol., Washington D.C.
- Laudelout H., Lambert R., Fripiat L. L. & Pham M. L.** (1974) Effet de la temperature sur la vitesse d'oxydation de l'ammonium en nitrate par des cultures mixtes de nitrifiants. *Ann. Microbiol. (Inst. Pasteur)* **127A**, 367-382.
- Lawrence A.W. and M<sup>c</sup>Carty P.L.** (1970) Unified basis for biological treatment design and operation. *J. Sanitary Eng. Div. ASCE* 757-779.
- Liebeskind M., Schapers D., Bornemann C., Brands E., Freund M., and Rolfs T.** (1996) Parameter determination and model fitting – two approaches for modeling process in wastewater treatment plants. *Water Sci. Technol.* **34**, 5-6, 27.
- Mauret M., Paul E., Costes C.P., Maurette M.T., and Baptiste P.** (1996) Application of experimental research methodology to the study of nitrification in mixed culture. *Water Sci. Technol.* **34**, 245.
- Mines R.O. Jr. and Sherrard J. H.** (1999) Temperature Interactions In The Activated Sludge Process *J. Environ. Sci. Health*, **A34**, 2, 329-340.

- Mobarry B.K., Wagner M., Urbain V., Rittmann B., and Stahl D.A.** (1996) Phylogenetic probes for analyzing abundance and spatial organization of nitrifying bacteria. *App. Environ. Microbiol.* **62**, 2156-2162.
- Monod J.** (1942) Researches sur la croissance des cultures. *Hermann and Cie, Paris.*
- Monod J.** (1949) The growth of bacterial cultures. *The Annual Review of Microbiology*, **3**, 371-394.
- Moore A.M.** (1986) Temperature and moisture dependence of decomposition rates of hardwood and coniferous leaf litter. *Soil Biol. Biochem.* **18**, 427-435.
- O'Kelley J.C.** (1970) Characterization of the particulate nitrite oxidase and its component activities from the chemoautotroph *Nitrobacter agilis*. *Biochem. Biophys. Acta.*, **205**, 409.
- Pano A. and Middlebrooks E.J.** (1983) Kinetics of carbon and ammonia nitrogen removal in RBCs. *J. Wat. Pollut. Control Fed.* **55**, 959-965.
- Painter H.A.** (1970) A Review of literature on inorganic nitrogen metabolism in microorganisms. *Water Research*, **4**, 6, 393-450.
- Painter H.A. and Loveless J.E.** (1983) Effect of temperature and pH value on the growth-rate constants of nitrifying bacteria in the activated sludge process. *Water Research*, **17**, 3, 237-248.
- Phelps E.B.** (1944) *Stream Sanitation*, John Wiley and Sons, Inc., New York, N.Y., 71-75.
- Poduska R.A. and Andrews J.F.** (1974) Dynamics of Nitrification in the Activated Sludge Process. *Presented at the 29<sup>th</sup> Industrial Waste Conference, Purdue University, Lafayette, Indiana.*



- Pommerening-Roser A., Rath G., and Koops G.P.** (1996) Phylogenetic diversity within the genus *Nitrosomonas*. *Syst. Appl. Microbiol.* **1**, 344-351.
- Prosser J.L.** (1989) Autotrophic nitrification in bacteria. *Adv. Microb. Physiol.* **30**, 125-181.
- Quinlan A. V.** (1980) The thermal sensitivity of nitrification as a function of the concentration of nitrogen substrate. *Water Research* **14**, 1501-1507.
- Quirk T.P. and Eckenfelder W.W.** (1986) Active mass in activated sludge analysis and design. *J. WPCF* **58**, 932-936.
- Randall C.W.** (1998) Activated sludge: latest developments and a look into the future. *J.CIWEM* **12**, p375-383.
- Silyn-Roberts G. and Lewis G.** (2001) In situ analysis of *Nitrosomonas* spp. In wastewater treatment wetland biofilms. *Water Research* **35**, 2731-2739.
- Sharma, B. & Ahlert, R.C.** (1976) Nitrification and nitrogen removal. *Wat. Res.* **11**, 897-925.
- Shieh W. K. & LaMotta E. J.** (1979) Effect of initial substrate concentration on the rate of nitrification in a batch experiment. *Biotech. Bioeng.* **21**, 201-211.
- Sozen S., Orhon D., San H.A.** (1996) A new approach for the evaluation of the maximum specific growth rate in nitrification. *Wat. Res.* **30**, 7, 1661-1669.
- Shuler M.L. and Kargi F.** (1992) *Bioprocess engineering: basic concepts* Prentice-Hall, Inc.
- Suzuki I.** (1974) Ammonia or ammonium ions as substrate for oxidation by *Nitrosomonas europaea* cells and extracts. *Journal of Bacteriology*, **120**, 556.

- Tchobanoglous G. and Schroeder, E.D.** (1985) "Water Quality: Characteristics, Modeling and Modification." Addison-Wesley Publishing; Reading, MA.
- Tendaj M., Reinius L.G., Hultgren J.** (1992) Some observations on nitrification and denitrification following full-scale trials at Henriksdal and Bromma sewage treatment plants in Stockholm. *Wat. Sci. Tech.* **25**, 4-5, 195-202.
- U.S. Environmental Protection Agency** (1993) *Process design manual for nitrogen control. Prepared for the Office of Technology Transfer of the USEPA, Brown and Caldwell*, Walnut Creek, California.
- Viessman and Hammer**, *Water Supply and Pollution Control*, 5<sup>th</sup> ed., Harper Collins College Publishers.
- Wagner M., Amann R., Lemmer H., and Schleifer K.H.** (1993) Probing activated sludge with *Proteobacteria*-specific oligonucleotides: inadequacy of culture-dependent methods for describing microbial community structure. *Appl. Environ. Microbiol.* **59**, 1520-1525.
- Wight J.R., Skiles J.W.** (1987) SPUR; Simulation of production and utilization of rangelands. Documentation and user guide. USDA-ARS pub. No. 63.
- Woese C.R., Weisburg W.G., Paster B.J., Hahn C.M., Tanner R.S., Kreig N.R., Koops H.P., Harms H., and E. Stackebrandt.** (1984) The phylogeny of purple bacteria, the beta subdivision. *Syst. Appl. Microbiol.* **5**, 327-336.
- Wong Chong G. M., and Loehr R.C.** (1975) The kinetics of microbial nitrification. *Water Res.* **9**, 1099.
- Yantarasri T., Garcia A. III, and Brune D.** (1992) Thermodynamic model of nitrification kinetics. *J. of Env. Eng.* **118**, 4.

## **Appendices**

## Appendix A. Full Scale Plant Data

Time	SRT	MLSS	Inf. BOD	Eff. BOD	Inf. NH4+	Eff. NH4+	SVI	F/M BOD5/MLSS/ HRT
1	11.6	2180	426	11	15.6	1.1	45.9	1.367
2	20.7	2350	461	6	17.2	1.9	42.6	1.372
3	16.4	2340	376	8	23.6	1	64.1	1.124
4	2.9	2860	528	3	21.4	0.7	49.0	1.291
5		3140	317	4	16.8	0.6	47.8	0.706
6		2950	376	12	13.3	0.4	54.2	0.891
7	1.8	2610	301	4	16.7	0.7	61.3	0.806
8	2.3	3270	202	2	17	0.5	61.2	0.432
9	2.0	3100	465	5	17.9	1.5	48.4	1.049
10	2.7	3160	282	6	19.4	3.9	50.6	0.624
11	2.8	3110	328	6	23.9	2.2	61.1	0.738
12		3530	230	3	21.4	0.9	56.7	0.456
13		2830	200	4	8.8	0.2	45.9	0.494
14	2.8	2970	337	4	19.8	2.1	57.2	0.793
15	3.5	4360	241	2	25.5	3.1	27.5	0.387
16	1.2	2020	252	4	24.5	3.8	74.3	0.872
17	2.0	2540	191	4	24	6.1	59.1	0.526
18	2.9	3060	304	2	23.5	1.6	49.0	0.695
19		3240	234	5	19.6	1.9	49.4	0.505
20		8860	176	3	13.9	0.2	9.0	0.139
21	2.7	3150	414	2	35	0.9	60.3	0.919
22	3.1	4530	514	1	32.3	4	28.7	0.793
23	3.4	5480	322	3	26.4	3.8	23.7	0.411
24	2.7	6110	218	3	23.3	3.1	19.6	0.250
25	1.0	2230	234	5	24.9	2.9	31.4	0.734
26		3810	247	4	20.3	2.1	39.4	0.453
27		6295	250	4	16	0.2	34.9	0.278
28	1.7	2670	388	3	0	0	52.4	1.016
29	1.6	3060	286	3	24.8	2.5	39.2	0.654
30	2.0	3900	330	2	29.8	4.8	38.5	0.592
31	2.2	4930	309	2	19.9	4	32.5	0.438
32	1.7	4330	291	15	22.1	2.1	34.6	0.470
33		4840	206	4	19.9	2.6	43.4	0.298
34		5850	216	3	13.6	0.7	46.2	0.258
35		5100	301	2	14.2	0.2	51.0	0.413
36		5310	269	6	12.3	0.9	13.2	0.354
37		7660	247	2	17.3	1	9.1	0.225
38		4630	268	15	17.5	1.5	56.2	0.405
39		3460	264	3	18.9	1.5	46.2	0.534
40		3930	215	4	36	1.1	50.9	0.383
41		2930	234	6	13.2	3	54.6	0.558
42	5.1	4460	313	2	18.3	0.3	29.1	0.491
43	56.9	3790	203	5	13.6	0.7	79.2	0.375
44	2.2	3120	575	6	18.2	0.9	48.1	1.289
45	2.0	3220	636	2	17.3	0.7	87.0	1.381
46	1.4	2010	392	4	23.7	1.8	99.5	1.364
47		3240	260	2	14.1	0.2	86.4	0.561
48		3270	287	2	14.6	0.2	76.5	0.614
49		3180	228	4	19.2	0.2	94.3	0.501
50		3270	227	4	18.7	0.2	116.2	0.485
51	2.0	3100	453	4	19	0.2	112.9	1.022

52	2.5	3480	194	2	18.5	1.1	63.2	0.390
53	2.3	3480	253	2	16.8	1.5	57.5	0.508
54	1.8	3040	456	4	20.4	1.7	55.9	1.049
55	2.1	3040	497	3	23.4	1.3	69.1	1.143
56		3370	285	4	17.2	0.4	74.2	0.591
57		3150	246	3	13.9	0.2	66.7	0.546
58	2.4	3070	233	4	20.3	0.2	81.4	0.531
59	3.0	3470	230	2	16.7	0.9	69.2	0.464
60	1.6	2460	523	2	18.5	0.2	97.6	1.487
61	2.1	2920	304	4	20.2	1	85.6	0.728
62	2.8	3460	552	4	20.9	0.6	106.9	1.116
63		3540	292	4	19.2	0.8	87.6	0.577
64	2.3	3040	450	5	21.7	0.8	115.1	1.035
65	3.0	3690	424	3	20	0.7	130.1	0.804
66	2.9	3870	352	5	20.2	3.1	113.7	0.636
67	2.1	3220	418	2	22.3	3.7	102.5	0.908
68		4200	351	4	21.3	2.7	107.1	0.584
69		3850	299	6	13.7	0.2	90.9	0.543
70	1.9	2190	692	2	18.9	0.5	255.7	2.210
71	5.1	6360	6200	2	17.7	0.9	26.7	6.817
72	2.6	2990	2810	5	15	2	90.3	6.572
73	1.7	2200	2610	4	15.8	3.3	90.9	8.296
74	2.9	4080	2890	3	20.5	0.9	39.2	4.953
75		3070	2730	5	24.2	2.7	42.3	6.219
76		3130	2850	6	16.1	1	63.9	6.367
77	1.9	2770	3940	4	23	2	65.0	9.947
78	1.5	1920	3970	3	16.4	0.5	83.3	14.459
79	2.4	3310	299	4	17	0.9	69.5	0.632
80	2.2	2420	371	6	22.1	2.6	74.4	1.072
81	2.2	2650	366	4	21.8	3.4	75.5	0.966
82		3180	212	3	19	1.5	66.0	0.466
83		3480	250	3	11.6	0.6	57.5	0.502
84	2.3	2470	353	3	25	1.4	68.8	0.999
85	3.1	3600	710	5	21.4	5.9	69.4	1.379
86	2.3	2950	578	4	17.7	5	57.6	1.370
87	2.6	3440	701	2	24.1	1.2	66.9	1.425
88		2180	362	2	18.2	0.2	64.2	1.161
89		3080	213	5	16.8	0.2	64.9	0.484
90		2680	271	4	19.8	0.2	56.0	0.707
91		2860	679	4	21.7	1.5	66.4	1.660
92	3.8	3340	552	3	18.4	1.6	80.8	1.156
93	2.5	2720	583	1	21.1	1.1	80.9	1.499
94	2.4	2310	363	5	25.9	3.2	60.6	1.099
95		3600	311	5	14.7	1.4	75.0	0.604
96		3560	366	5	15.1	0.2	75.8	0.719
97		2550	401	1	12.8	0.2	82.4	1.100
98	6.9	6250	620	3	16.5	0.8	25.6	0.694
99	3.1	3330	509	4	11	0.4	87.1	1.069
100	3.3	3620	381	4	13.5	0.3	82.9	0.736
101	6.3	3000	359	4	16.6	0.7	86.7	0.837
102	2.7	3420	302	3	15.5	0.2	78.9	0.618
103		3440	203	4	15.5	0.6	78.5	0.413
104		2710	447	20	6.4	2.1	92.3	1.153
105	2.3	2710	269	23	6.7	2.1	88.6	0.694
106	3.1	3750	482	2	8	0.5	130.7	0.899
107	3.6	4350	519	4	14.9	1	50.6	0.834

108	1.9	3120	167	6	10.2	1.6	83.3	0.374
109	2.2	3000	269	6	12	1.8	83.3	0.627
110		3420	167	4	11.2	0.2	73.1	0.341
111		2580	231	7	10.9	0.2	89.1	0.626
112		2970	237	4	10.3	0.5	87.5	0.558
113	3.7	4120	210	3	14	1.7	65.5	0.356
114	2.1	2850	257	6	18.1	3.3	73.7	0.631
115	2.0	2620	259	3	15.8	2.1	80.2	0.691
116	2.5	3070	209	4	15.2	1.5	91.2	0.476
117		3800	219	3	16.2	1.5	68.4	0.403
118		3110	221	2	14.4	0.6	74.0	0.497
119	2.4	2500	210	3	17.2	1.9	76.0	0.587
120	2.8	3240	184	3	22.7	2.2	92.6	0.397
121	2.5	2990	284	3	21.1	1	73.6	0.664
122	2.7	2730	251	4	17.4	5.4	73.3	0.643
123	3.4	3460	196	4	27	7.5	86.7	0.396
124		3310	220	4	15.2	0.9	129.9	0.465
125		2660	223	4	12.8	0.3	94.0	0.586
126	3.0	3840	239	1	17.2	0.4	65.1	0.435
127	8.9	2950	282	3	15.3	3.6	74.6	0.668
128	6.9	2800	209	5	18	2.6	67.9	0.522
129	6.1	3380	259	1	19.4	1.5	88.8	0.536
130	5.3	3810	288	1	18.7	0.5	81.4	0.529
131		3550	221	5	15.2	2.4	87.3	0.435
132		2880	208	5	12.1	0.3	86.8	0.505
133	4.0	3110	301	11	20.5	3.3	96.5	0.677
134	3.2	2470	803	3	22	2.1	85.0	2.273
135	4.0	2870	373	1	19	1.7	90.6	0.909
136	4.5	3110	232	3	19.2	7.2	90.0	0.522
137	3.4	2540	439	5	24.8	8.7	94.5	1.209
138		3130	261	4	12.3	0.7	86.3	0.583
139		2610	207	4	9.2	0.2	80.5	0.555
140	5.5	2980	197	5	10.9	0.3	97.3	0.462
141	4.7	4410	221	5	13.8	5.1	36.3	0.350
142	4.0	2930	274	0	16.3	7	68.3	0.654
143	3.9	2920	192	2	16.6	3.4	75.3	0.460
144	3.7	3220	261	3	9	1.2	80.7	0.567
145		3640	157	4	5	0.2	57.7	0.302
146		3640	181	12	5	0.2	82.4	0.348
147	5.1	2960	228	4	12.1	3.7	81.1	0.539
148	3.7	2280	253	4	13.1	1	65.8	0.776
149	4.3	2530	258	2	14.1	1.1	63.2	0.713
150	4.7	3940	231	4	15.2	5.3	78.7	0.410
151	4.0	3340	244	5	19.5	6.2	80.8	0.511
152		2890	210	13	14.9	2.5	69.2	0.508
153		2680	214	4	7.8	0.3	85.8	0.558
154		3030	187	6	15	2	92.4	0.432
155		2580	416	9	21.6	4.5	89.1	1.128
156	4.4	3140	242	3	16	8.8	60.5	0.539
157	3.5	2760	217	2	18.4	1.4	87.0	0.550
158	5.1	2770	256	6	19.9	0.2	75.8	0.646
159		3300	207	5	12.8	2.1	60.6	0.439
160		3330	196	6	10.6	0.7	63.1	0.412
161	3.1	2460	233	6	20.2	2.6	89.4	0.662
162	2.5	1840	339	5	13.6	11.1	65.2	1.288
163	4.9	2870	244	3	15.7	6.4	76.7	0.595

164	3.6	3320	423	6	16.4	11.8	57.2	0.891
165	3.7	3050	228	4	18.1	10	72.1	0.523
166		2310	216	4	5.8	0.2	64.9	0.654
167		2320	136	3	8.5	0.2	77.6	0.410
168	3.9	2490	193	4	10.6	1.9	68.3	0.542
169	2.4	2590	180	6	11.1	4.1	65.6	0.486
170	2.6	2920	200	4	14.1	3.7	71.9	0.479
171	1.8	1700	259	4	13.2	2.7	70.6	1.065
172	3.4	2850	195	2	10.5	0.9	70.2	0.478
173		3390	221	3	15.8	0.4	76.7	0.456
174		4780	192	7	9.9	0.4	43.9	0.281
175	2.1	2330	120	10	4	1.1	77.3	0.360
176	3.5	3020	296	2	11.2	0.2	66.2	0.685
177	3.5	3030	208	2	5.1	0.2	66.0	0.480
178	3.1	2480	154	4	9.6	1.8	76.6	0.434
179	3.9	3340	320	2	16.4	6.8	65.9	0.670
180		3650	557	3	12.2	0.2	68.5	1.067
181		3630	260	3	9.5	0.2	66.1	0.501
182	4.2	3200	228	6	8.4	0.2	53.1	0.498
183	4.4	3810	516	2	13.6	2.3	34.1	0.947
184	2.8	2700	708	5	13.4	2.3	70.4	1.834
185	3.6	2690	691	1	9	0.9	63.2	1.796
186	4.7	2990	282	5	12.8	1.4	80.3	0.660
187		3180	263	4	14.7	2	78.6	0.578
188		3500	212	4	6.8	0.9	57.1	0.424
189	2.2	3220	135	31	4.2	3.8	65.2	0.293
190	2.2	2410	148	12	3	0.7	83.0	0.429
191	1.3	2420	163	7	5	0.3	62.0	0.471
192	1.6	2420	99	8	3.8	1.5	78.5	0.286
193	2.4	3180	303	5	12.4	1.2	72.3	0.666
194		2830	167	4	6.5	0.2	70.7	0.413
195		2800	162	4	9.1	1.2	60.7	0.405
196	2.3	2630	517	3	11.3	0.6	76.0	1.375
197	2.2	3090	204	2	10.2	2.3	80.9	0.462
198	2.2	2610	287	8	10.9	3	76.6	0.769
199	1.9	2620	208	4	5.8	0.9	76.3	0.555
200	2.4	3040	133	2	10.8	0.7	88.8	0.306
201		2530	119	4	9.5	1.1	75.1	0.329
202		2320	129	3	7.1	0.2	90.5	0.389
203	2.2	2650	185	4	11.5	0.8	94.3	0.488
204	2.2	2840	118	5	11.6	5	88.0	0.291
205	2.5	3080	539	3	13.8	7.3	94.2	1.224
206	2.7	3750	284	6	13.6	7.6	88.0	0.530
207		4200	177	2	15	4.5	107.1	0.295
208		3270	323	2	6.3	2	100.9	0.691
209		2780	202	3	12.6	0.2	107.9	0.508
210		3130	360	4	8.8	0.2	92.7	0.804
211		2610	85	9	7	2.9	92.0	0.228
212	1.9	2790	335	11	11.5	2.7	100.4	0.840
213	2.3	2610	262	2	13.7	0.4	76.6	0.702
	2.2							

## Appendix B. Bench Scale Data

Time	5-Day MLSS	Inf. COD	Eff. COD	Inf. NH4+	Eff. NH4+	SVI	F/M mg COD/mg MLSS.day
1	880		62.1	24	0.1		
2	1300			15.7	0.1	61.5	
3	1440		41.7	16.1	0.1	59.0	
4	1390		76	15.8	0.1	61.2	
5	1170	116.3	30	19.2	0.12	64.1	0.272
6	1220	204	43	19.2	0.12	61.5	0.458
7	1110	181.1	42	21.9	0.1	73.9	0.447
8	980	155.3	22	22.1	0.1	83.7	0.434
9	980	155.3	38.3	22.2	0.1	173.5	0.434
10	950	275.7	37.4	19.7	0.1	578.9	0.795
11	930	243.2		20.2	0.1	946.2	0.716
12	850	252.9		19	0.1	1141.2	0.815
13	840	246.4		19.4	0.1	1188.1	0.804
14	870	272.4		21.1	0.1	1092.0	0.858
15	730	217.1	72	20.1	0.1	1212.3	0.815
16	810			22.2	0.1	1123.5	
17	870	249.7		21.5	0.1	1023.0	0.786
18	860	477.4	82	24	0.1	970.9	1.521
19	970	483.9		23.6	0.1	835.1	1.367
20	1010			24.4	0.1	752.5	
21	840	236.6	144.3	23.3	0.1	1071.4	0.772
22	1070	366.8	69.1	23.9	0.1	831.8	0.939
23	1230	623.8		48.8	17.7	512.2	1.389
24	1780	636		49.2	31.6	387.6	0.979
25	1970					355.3	
26	2270	867.8		45.4	22.6	383.3	1.047
27	2340	649.9		54.3	13.3	363.2	0.761
28	2760	496.9	118.4	40	0.2	322.1	0.493
29	3000	155.3	104.3	39.5	0.1	278.3	0.142
30	2980	383.1	78	33.8	0.1	310.4	0.352
31	2960	321	65.7	34	0.1	250.0	0.297
32	2940		85.4			265.3	
33	2740	285.4	65.2	35.5	5.81	226.3	0.285
34	3060	539.3	79	40	0.1	178.1	0.483
35	3346		105	39.6	0.1	257.0	
36	2960		111.7	39	0.1	206.1	
37	3120	274.6	60	27.9	0.1	169.9	0.241
38	2840					162.0	
39	2460	173.1	51.7	23.6	0.1	170.7	0.193
40	2340	170	29.4	22.8	0.1	149.6	0.199
41	2380					175.6	
42	2240	190.6	38.2	23.5	0.1	200.9	
43	1920	220	57.3	24.7	0.1	234.4	
44	1740	573.1		24.4	0.1	252.9	0.902
45	1520	246.9		24	0.1	289.5	0.445
46	1540	157		23.3	0.1	344.2	0.279
47	1580	607.6				489.2	1.054
48	1340	279.5	61.0209			649.3	
49	1240			25.4	0.1	564.5	
50	1340			34.2	0.1	522.4	
51	1290	282.2	71.8464	30.2	0.1	465.1	0.599



52	1340	288.7	73.57848	31.9	0.1	425.4	0.590
53	1290	265.9	82.6719	29.7	0.1	347.3	0.565
54	1380					202.9	
55	1250	226.9	73.57848	29.3	0.1	80.0	0.497
56	1160	311.5	63.61902	18.8	0.1	38.8	0.736
57	1090	233.4	78.3417			36.7	0.587
58	1010					29.7	
59							
60	860	308	60.58788	20	0.12	34.9	0.981
61	780						
62	930	662	52.2			26.9	1.950
63	1050	228	55.3	19.5	0.1	28.6	0.595
64	1050	265	75	20.7	11.3	28.6	0.691
65	1220	222	137	19.8	15.5	32.8	0.499
66	1180	210	108.3	20.5	16.4	40.7	0.488
67	1240	754	84			96.8	1.666
68	1050	163	84.9	21.6	17	85.7	0.425
69	1300	376.2	52.6	22.2	17.9	96.2	0.793
70	1270	213	94.2	22.3	19.3	133.9	0.459
71	1330	222	63	24.4	20.6	169.2	0.457
72	1220	222	69.5	23.1	21.3	192.6	0.499
73	1080					277.8	
74	1010	397	63	22.7	21.1	267.3	1.077
75	1030	126	70.5	19.8	19.2		0.335
76	970					360.8	
77	880	213	85.5	17.7	16.4	272.7	0.663
78	950	163	91.6			368.4	
79	970			19.9	17.6	345.4	0.460
80	1740			19.1	18.5	136.8	
81	2130					79.8	
82	2200	330	71.9	19.1	17.3	70.5	0.411
83	2000					65.0	
84	1870	243	75.2	20.1	18.3	53.5	0.356
85	1620	166.9	70.5	20	19.2	49.4	0.282
86	1740	185	82.2	21.8	18.7		
87	1480	256.1	93	23.9	20.1		
88							
89							
90							
91	1320	298	105	23	20.5	75.8	
92	1460	275.3	66.8	21.9	20.8	68.5	
93	1640	242	79.2	20.3	21.1	57.9	
94	1480	210	47.8	22.6	21.7	64.2	
95	1320	189.5	42.25	26.7	21.6	64.4	
96							
97	1200					100.0	
98	1240	203.8	70.6	28.6	23.5	141.1	
99	1400	259.6	86.1	27.3	24.6	178.6	
100	1500			26.3	23.5	133.3	0.372
101	1360			15.1	16.4	216.9	0.523
102	1320			14	15.3	185.6	
103	1040	220.2		10.5	11.9	177.9	0.580
104	960			9.9	10.2	130.2	
105	820	105.2	52.4	10	10.2	103.7	0.351
106	760			9.7	12.7	98.7	
107	620					72.6	
108	640	101.9	49.5	8.2	7.4	62.5	0.436

109	620	121.6	33.3	9.1	7.5	61.3	0.537
110	560					53.6	
111	400			12.3	6.3	62.5	
112	410	170.9	64.9	14	8.9	61.0	1.142
113	510	213.6	58.2	15.7	11.1	49.0	1.147
114	530	177.5	57.4	14.8	11.7	56.6	0.918
115	540	630.9	35.6	15.8	9.7	46.3	3.201
116	640	371.3	48.6	15.6	9.5	46.9	1.589
117	680			16.1	8.8	55.9	0.675
118	720			17	8.8	55.6	1.038
119	770	167.6	52	15.9	8.6	58.4	
120	770	272.7	52.4			51.9	
121	960			19.8	7.1	60.4	
122	1040		50	20.7	9.1	65.4	
123	1140			21.5	9.5	61.4	
124	1320					64.4	
125	1280					66.4	
126	1440	305.6	77.3	26.1	8.9	69.4	0.581
127	1420	420.6	76.1	25	7.6	70.4	0.811
128	1480	368.2	81.5	34.1	23.7	74.3	
129	1440	408.7	74.9	28.2	8.1	79.9	
130	1420	368	55.3	28.5	4.27	83.1	
131	1320					90.9	
132	1240					88.7	
133	1200		47	23.1	0.11	79.2	
134	1240		60.4	22.5	0.54	76.6	
135	1060	368		22.4	0.1	75.5	0.951
136	1020	343	105	24.2	2.47	73.5	
137	1040	305	100.5	23.5	5	67.3	0.803
138	860					58.1	
139	940					62.8	
140	850	285	93.2	22.4	4.21	58.8	
141	700	174.2	58.7	22.5	1.58	54.3	
142	540	49.3		11.6	0.1	55.6	0.250
143	550			11.7	0.1	54.5	
144	640					39.1	
145	370					67.6	
146	500					50.0	
147	500	75.6	21	16.1	0.1	50.0	0.414
148	570	105.2	28	15.3	0.1	49.1	0.506
149	570	141.3	35			49.1	0.679
150	690			16.1	0.198	40.6	
151	530	135.2	41.4	14.1	0.1	47.2	
152	690					43.5	
153	720					34.7	
154	650	140.6	50.5	16.1	0.1	38.5	
155	680	172.8	68.8	16.1	0.1	41.2	
156	710					39.4	
157	850	197.2	132.8	19.8	0.1	49.4	0.064
158	850			20.2	0.1	47.1	
159	870					46.0	
160	790	180.8	122.3	20.1	0.1	50.6	0.627
161	900	134.5	35.4	19.8	0.1	50.0	0.409
162	960	190.6	28.6			57.3	0.544
163	1040	182.4	44.6	18.9	0.1	56.7	
164		177.5		17.5	0.1		

165	940	154.5	164.1				0.450
166	1030	161	72.1	21.4	0.1	58.3	0.428
167	900			21.4	0.1	66.7	
168	900	151	74.3			77.8	0.460
169	860			20.8	0.1	80.2	
170	900	197.2	36.4			66.7	0.600
171	860			20.6	0.1	69.8	
172	900			19	0.1	66.7	
173	810					80.2	
174	690	190.6		26.2	0.1	72.5	0.757
175	660					68.2	
176	840			26.1	0.1	77.4	
177	898					75.6	
178	780			19.5	0.1	102.6	
179	670					59.7	
180	610			21.1	0.12	49.2	
181	560					53.6	
182	520	216.9	97.7	16.9	0.1	53.8	1.143
183	480					52.1	
184	470			18	0.11	53.2	
185	450					44.4	
186	450			13.4	0.1	44.4	
187	440	210.3					1.309
188	470					53.2	
189	430	197.2		12.8	0.2	58.1	1.256
190	640	210.3		12.6	0.15	46.9	0.900
191	720			12.5	0.1	48.6	
192	630					47.6	
193	520			12.9	0.16	57.7	
194	390					64.1	
195	470					53.2	
196	520			13.5	0.1	48.1	
197	650			13.1	0.1	38.5	
198	870			13.2	0.1	34.5	
199	840					35.7	
200	820			12.9	0.1	34.1	
201	900					33.3	
202							
203	720			11.5	0.1	34.7	
204	740					33.8	
205	630					39.7	
206	630					31.7	
207	610			9.1	1.85	32.8	
208	640					39.1	
209	670					37.3	
210	800					37.5	
211	780					38.5	
212							
213	640					0.0	

## **Vita**

Jack McClendon Carley was born in Knoxville, Tennessee on September 19, 1974. He attended the public system of schools in Knox County, where he graduated from Bearden High School in May 1992. He entered the University of Tennessee-Knoxville in August 1992, receiving a Bachelor of Agriculture in Plant and Soil Science in December 1996. Mr. Carley entered the Master's program in Environmental Engineering as a full time student in August of 1998. Working as a graduate research assistant, he officially received his Master of Science Degree in Environmental Engineering in April 2003.